

# 東京帝國大學紀要

THE

# JOURNAL

OF THE

# COLLEGE OF SCIENCE, IMPERIAL UNIVERSITY OF TŌKYŌ, JAPAN.

VOL. XIII.

東京帝國大學印行 PUBLISHED BY THE UNIVERSITY.

TÖKYÖ, JAPAN.

1900-1901.

MEIJI XXXIII-XXXIV.

# CONTRIBE

e 2na, 1900.	
Notes on the Geology of Art of Smdent Isles of Taiwan.  By B. Koto, Ph. D. Karaka for Shi, Professor of Geology,  Science College, Imperiation ity, Tokyo. (With Plates  I-V)	1
Change of Volume and of I earth of Iron, Steel, and Nickel	
Ovoids by Magnetizah (1982) (1. NAGAOKA, Rigakuhakushi,	
Professor of Applied Ma and K. Honda, Rigakushi,	
Post-graduate in Physics (her Plates VI & VII)	57
Combined Effect of Longitudinal and Circular Magnetizations	.,,
on the Dimensions of them, Steel and Nickel Tubes.	
By K. Honda, Rigalush a last raduate in Physics. (With	
Plates VIII & IX)	77
Studien uber die Anpassangsfahigt bit einiger Infusorien an	• •
concentrirte Losungen. Vis Yasuda, Rigakushi, Pro-	
fessor der Naturgeschicht aus ir Zweiten Hochschule zu	
Sendai. (Hierzu Tafel	101
Ueber die Wachsthumsbesone einiger Algen und	101
Pilze durch chemische 1601 Von N. Oso, Rigakushi.	
(Hierzu Tafel X-XIII)	141
(11101410 10fct A-A111)	1.11
Pt. II. Publ. 1 ly 25th, 1900.	
Ammonium Amidosulphite Divers and M. Ogawa.	
Ammonium Amidosulphite Divers and M. Ogawa. Imperial University, Tol	187
Products of heating Amnagine Sulphites, Thiosulphate,	
and Trithionate. By Mark and M. Ogawa, Imperial	
University, Tokyo	201
Pottassium Nitrito-hydroxynade alphates and the Non-	
existence of Dihydran arrive Derivatives. By E.	
DIVERS, M.D., D. Sc., F eritus Prof., and T. HAGA.	
D. Se., F. C. S., Professo Imperial University	211

A. 7- 1

tification and Constitution of Fremy's Sulphazotised Salts of Pottasium, his Sulphazate, Sulphazite, etc. By	
E. Divers, M.D., D. Sc., F. R. S., Emeritus Prof., and T. Haga,	
	225
On a Specimen of a Gigantic Hydroid, Branchiocerianthus	
imperator Allman). found in the Sagami Sea. By M.	
Miyajima, Rigakushi, Science College, Imperial University,	
Tokyo. (With Plates XIV & XV)	235
Mutual Relations between Torsion and Magnetization in Iron	
and Nickel Wires. By H. NAGAOKA, Rigakuhakushi, Pro-	
fessor of Applied mathematics, and K. Honda, Rigakushi,	
Post-graduate in Physics. (With Plates XVI)	263
The Interaction between Sulphites and Nitrities. By E.	
DIVERS, M. D., D. Sc., F. R. S., Emeritis Prof., and T. HAGA,	
D. Sc. F. C. S., Professor, Tokyo Imperal University	281
Pt. III. Published Dec. 28th, 1900.	
Contributions to the Morphology of Cyclostomata. II.—The	
Development of Pronephros and Segmental Duct in Petromyzon.	
By S. Hatta, Professor in the College of Peers, Tokyo. (With	
Plates $\widehat{XVII}$ - $XXI$ )	311
Beiträge zur Wachstumsgeschiehte der Bambusgewachse.	011
Von K. Shibata, Rigakushi. (Mit Tafeln XXII-XXIV)	427
Decomposition of Hydroxyamidosulphates by Copper	
Sulphate. By E. Divers, M. D., D. Sc., F. R. S., Emeritus	
Prof., and T. Haga, D. Sc., F. C. S., Professor, Tokyo Imperial	
University	497
Pt. IV. Published Oct. 5th, 1901.	
Observations on the Development, Structure and Metamor-	
phosis of Actinotrocha. Iwaji Ikeda, Riyakushi. (With	
Plates XXV-XXX)	508

# Publish mmittee.

- Prof. K. Mitsukuri, Ph. I wakushi, Director of the College (ex officio).
- Prof. B. Kotô, Ph. D., Riga Forter
- Prof. T. Haga, Rigakuhakusta.
- Prof. S. Watasé, Ph. D., I hi.

All communications relatin \_\_\_\_\_ ournal should be addressed to the \_\_\_\_\_ Director \_\_\_\_\_ coalege of Science.

#### CORRIGENDA.

```
for Gi-ô-tô
                                                     read
                                                           Gio-ô-tô-
Page
              12th line,
                                                     read
                                                           Cholnokv.
       6,
                                    Cholnecky
              the last line,
                               for
                                   there lies
                                                     read
                                                            the relics.
                               for
              18th line,
      18,
                                                            bastite.
                                                     read
                               for basite
              the last line,
      21,
 17
                               for
                                    crystals
                                                            erystal.
      \bar{2}8,
                                                     read
              9th line,
                                                           Bücking
                                                     read
                                for
                                    Büking
      29,
              13th line,
                              delete the word 'macroscopically.'
      42,
              2nd line,
                                                           ' leached or percolated.'
                                    'leached'
              14th line,
                               for
                                                     read
      45,
                                                            pyrite.
                                                     read
                               for
                                    pyrites
      53,
              24th line,
 ,,
                               for
                                    granual
                                                            granular.
              20th line,
                                                     read
                                                     read
                                                           quadrant.
 " Explanation Pts. I & II,
                               for
                                    octant
```

between the, geologically neglected, south-east coast of China and Taiwan, the expanse of sea is studded with a great number of islands, collectively called the Hôko or Pescadores Group. It consists of islands, islets and rocks, great or small, altogether numbering 57, besides countless hidden rocks under the water. The waterway on the continental side of the Pescadores is the shallow Fokien Strait, only a hundred miles wide, and on the Taiwan side, is the still narrower Hôko Channel,—the only passages which allow free communication to the waters of the de-

Publishing Committee.

# Notes on the Geology of the Dependent Isles of Taiwan.

By

B. Kotô, Ph. D. Rigakuhakushi,Professor of Geology, Science College, Imperial University, Tôkyō.

With Plates I-V.

## THE HÔKO GROUP (PESCADORES).

### I. Introductory.

Between the, geologically neglected, south-east coast of China and Taiwan, the expanse of sea is studded with a great number of islands, collectively called the Hôko or Pescadores Group. It consists of islands, islets and rocks, great or small, altogether numbering 57, besides countless hidden rocks under the water. The waterway on the continental side of the Pescadores is the shallow Fokien Strait, only a hundred miles wide, and on the Taiwan side, is the still narrower Hôko Channel,—the only passages which allow free communication to the waters of the de-

pressions of the North and South China Seas. The region is alternately subjected to strong ebbs and floods through the

influence of the branch currents of the swift Kuro-shiwo from the north and south, creating foamy and turbulent waves, in conjunctin with the steadily blowing heavy north-easters,—the dread of coasting navigators for ship-wrecks and other deplorable accidents.

I have not yet had opportunity to learn by my own inspection the geology of the Pescadores Group; but through the kindness of Messrs. Y. Saitô and

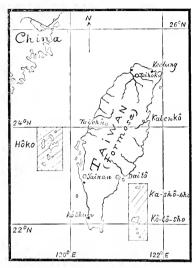


Fig. 1.—Index map of Taiwan to show the position of the islands described.

T. Tada, I have obtained about forty specimens of rocks, which no doubt fairly represent the types that build up the crust of the islands. In anticipation of a fuller report by Prof. Yokoyama, who has made the islands the subject of his special study, I may give here brief notes on the descriptions of rocks and the inference drawn as to the probable geologic structure of this interesting volcanic group.

The islands are, broadly speaking, distributed within an elliptical space. On the north of the Tropic of Cancer lie mainly the larger islands which are arranged after the manner of Santorin. They resemble the latter not merely in general outlines, but they owe their very existence to the same cause; both are of volcanic origin. These Santorin-like islands are Gio-ô, Hôko, Hakusha, and Chû-don, the latter three fuse together, especially during low tide, into one mass with the intervening

coral-reefs which stretch from one island to the other, making the shape very much like Thera. The single island of Gio-ô, then, corresponds in shape and position to that of Therasia. Here, however, we look in vain for the active centre of Kaimenis of Santorin. Taking into account the general distribution of the above-mentioned islands, and also the bathometrical condition, which the chart, Plate IV plainly shows, it is likely that they form an independent centre of effusion, in contrast to the Southern group (the Rover group), from which this Northern is separated by the Rover Channel, though both sit upon the eastern end of the so-called Formosa Bank, which stretches out hither from the coast of Fokien. The same type of topography seems to prevail throughout the whole group. It is simple, monotonous, flattopped and low; the highest prominence scarcely exceeds 56 m. (located at the south-west point of (Sio-ô), and the land can only be recognised from the sea within few miles. The islands consequently are wanting in wind-protected harbours, being constantly exposed to the north-east stiff gales during full three-quarters of a year. The land surface is bare, desolate and barren, being entirely destitute of green covering, due, it is said, mainly to the savage violence of the wind, against which even hardy shrubs can not maintain their footing.

The rain-fall, which the south winds occasionally brings thither during the summer season, is soaked up as soon as it falls on the eraggy ground; and there are searcely any rivulets that properly deserve the name. The erosive actions of running water thus become totally suspended, and valleys and dales are searcely to be seen in the interior, but only the butte-like table-land capped with the Basalt-sheet. The deflation alone is instrumental in modelling the topography, and here we

have a quasi-desert, and not an oasis, amidst the green island-world of South-eastern Asia.

Forty of Mr. Tada's specimens of rocks, on which I base my petrographical descriptions in the present paper, were collected from the following islands:—

- 1) Hôko island, the largest of the whole group.
- 2) Haku-sha-tô, 1) lying north to the foregoing.
- 3) Impai-sho.
- 4) Chô-sho, the eastern neighbour of Hakusha-tô.
- 5) Kippai (Bird Island of English Admirality chart), the northernmost of the whole group.
- 6) Gi-ô-tô (Fisher Island), west of Hôko-tô.
- 7) Hattô-sho, lying farther to the south of the main group.

In addition to these, I have received lately a few specimens collected by Mr. Y. Saitô.

<sup>1)</sup> The words 'tô' and 'sho' recurs frequently in the geographical name of Taiwan, the former signifying an island, the latter an islet or rock.

# II. Stratigraphical Characteristics.

## HÔKO ISLAND.

Hôko or Tai-san-sho<sup>1)</sup> is the largest among the forty-seven islands of the Hôko group, having an area of 62.7 square kilométres. Its general outline is k-shaped, curving in at three points in the coves, Fûkibi,<sup>2)</sup> Giû-bo-ken,<sup>3)</sup> and Kôtei.<sup>4)</sup> The relief is simple, low and flat-topped, the maximal elevation being Mount Tai-bu,<sup>5)</sup> located nearly at the centre, with a height of only 48 m. The coast is cliffy, interrupted often by sandy flats fringed with coral reefs.

Mr. Y. Saitô has geologically reconnoitered the principal islands of the group during last winter, and has kindly placed at my disposal the written account of his observations, which I am here following in its main points.

The island is essentially composed of the Tertiary Basalls, of which three different flows, poured out after long intervals, are well marked by the intervening tufaceous sedimentaries of a considerable thickness. The topmost flow caps the surface of butte-like elevations, or makes the flows of extensive 'mesas,' the surface being covered with its cluvial products—a fine, ferruginous loam which gradually passes downwards into a blocky loam and then the massive lava. The flow is rather thin, and characteristically columnar. It is frequently wanting in some parts of the island.

In the irregularly formed strip of land—the Fükibi-Jiri<sup>6</sup>

<sup>1)</sup> Tai-san-sho (大田嶼), signifying 'great mountain islet,' is by no means literally true, though undoubtedly it is the largest of the whole Pescadores.

<sup>2)</sup> 楓櫃美 3) 牛母件 4) 港底 5) 大武山 6) 塒徑.

tongue, which projects out from Sei-shi-an¹ towards the citadel of Bakô, thus enclosing within it a safe harbour,—we see the second sheet of flow, beautifully exposed along the steep declivity all round the shore under the uppermost lava-flow, from which it is separated by a thin bed of tuffite. This is a most extensive and strong sheet, aggregating about 10 m. In its upper portion, the lava is porous, whitish, and much decomposed, while the lower portion is fresh and compact. It is the one which we usually see along the sea-shore on whose trappean floor the rollers break and recoil in tumultuous wayes.

The third is the lowest, consequently the oldest flow visible in the Pescadores, and frequently forms the floor of the coast, when the second sheet, already referred to, makes its appearance higher up the precipice. It is likewise doleritic and porous as in the above flow, and this Basalt is well seen at the environs of Jiri, already referred to, where it is underlaid by a meagre lignite-bearing bed. It rarely happens to come to the surface not because of its absence but that it is hidden under the level of sea.

Tertiary strata, often accompanied by lignite seams, occur inserted between the first and second flows, and also below the third sheet. An undeterminable cast of gasteropod together with an Area were secured by Saitô from the corresponding bed at Run (Lun) point in the Island of Gio-ô. The sure proofs of their being of the Tertiary age are not at hand; but from the analogy of the occurrences of Basalts in the neighbouring regions, I conjecture the sedimentaries, here referred to, to be of later Tertiary. According to Cholnecky<sup>2</sup>, two volcanic lines are said to be

<sup>1)</sup> 井仔垵

Vorläufiger Berichte über meine Forschungsreise in China.' Petermanns Mitth. 45, 1899, S. 8.

distinguished in Eastern Asia; the one has served for the welling out of an enormous quantity of Basalt in later Tertiary age, the other has given rise to the chains of modern (Andesitic) volcanoes. In the north of the Chang-pei-shan, in Korea, he announced recently the discovery of an extensive Basaltic mesa more than 60,000 square km., which extends from Mukden through Kirin to Ninguta, forming the water-shed of the Sungari River and the Tumen-kiang. I have been informed, verbally by Mr. Nishiwada, of the occurrence, outside of Manchuria, of a trappean plateau, of small extent, along the eastern water-shed of the Korean Peninsula, and the island of Quelpart; and Vénukoff<sup>n</sup> cites a number of localities where Basalts make their appearance on the plateau of Mongolia. Furthermore, the Basalts occur sporadically in Liau-tung, and Shang-tung as far down as Nanking, approximately in a straight line, and v. Richthofen2) brings the line in connection with the tectonic movement which has created the 'great plain' of China, and he assigns the age of this crustal movement to the Tertiary period. The Basalts of the Pescadores seem to me to be included in this petrographical province of Eastern Asia.

Since the beginning of the *Dilurial epoch*, a subaerial condition has prevailed over Hôko, as well as in all the islands of the whole group, and erosion and disintegration have been at work, thereby carrying off the greater part of the uppermost flow, and gradually diminishing the area of the islands, and finally reducing them to ruins, as we see at present. Consequently, no record is left of the deposit representing this period, unless we take for it the

<sup>1) &#</sup>x27;Les Roches basaltiques de la Mongolie,' Bulletin de la Société belge de Géologie, etc., tome II, p. 441.

<sup>2) &#</sup>x27;Shantung und seine Eingangspfort Kiantschou,' 1898, S. 66.

thin superficial covering of ferruginous loam which is in part at least the product of decay of Recent epoch, though a certain portion may have been defladed away and lost during dust-storms.

Along the shore free from escarpment, white sandy beaches stretch from one point to another. They are the Alluvial deposits, into whose composition enters a special element which we are not accustomed to see in our own coast. Nearly all round the island, coral reefs grow upon the Basaltic shelf, and the detritus derived from them is driven up to form low sand dunes, leaving behind them, if the coast-line is deeply indented, as it is in many places, muddy shallows filled with the residual clay of decomposed Basalt.

Such is the general outline of the geology of the Island, and of the rest of the group as well.

Looking more into the details, we find that at Bakô¹¹ Point, on which is situated the town of the same name, the second flow extends in a great sheet, covering all but a few points of elevation which are capped with there lies of the young columnar lava, being separated from it by a blue rock. The last is a fuller's earth, which is a bluish-grey, dull, compact mass of greasy lustre, splitting, when dry, into angular clods with sub-conchoidal fracture. It adheres to the tongue, and falls readily to a muddy state on placing in water, and is not plastic. Under the microscope, the whole mass consists of brownish, double-refracting particles, and seems to have been derived from the decomposition of a Basaltic glass. It crops out for a short distance, and on shore a poor bed of Tertiary lignite occurs associated with it.

<sup>1)</sup> 媽公.

The same state of things prevails throughout the tract southward as far as Sei-shi-an<sup>1</sup>, the surface being covered with thick ferruginous loam mixed up with Basaltic fragments, and the upper and middle flows coming in direct contact, distinguish-

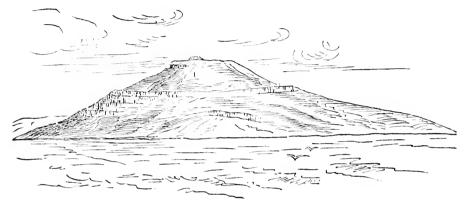


Fig. 2.—Isolated erosion hill Sha-bo-san, near Jiri, showing two upper flows with interbedded sedimentaties.\*

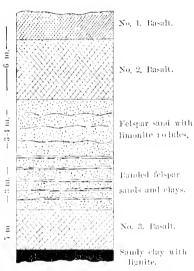
able only in the difference of structures. At the last-mentioned locality, a 'haul-over' of base-levelled middle flow, masked with coral sand, separates the tongue of land Jiri<sup>2</sup>, on which stands a Basaltic, hat-shaped Sha-bô-san<sup>3</sup>, 47 m. high (Fig. 2).

A good section may be seen along the shore, west of Jiri, as is shown in Fig. 3. The columnar, upper (No. 1), and doleritic, porous middle flows (No. 2), aggregating about 6 m., cap the cliff, 20 feet high. That the two flows are separated by long time intervals can be clearly shown elsewhere (Fig. 2) by a bed inserted between them. I may cite the case of a lignite bed at Bakô, occurring in company with fuller's earth. Another instance may be given of it just east of Jiri, where an ash bed makes its appearance. This ash bed is a fine, greyish-white, pulverent

<sup>1)</sup> 井仔坊 2) 塘裡 3) 紗帽山。

<sup>&</sup>quot;All the figures in the following wood-cuts, not otherwise mentioned, are originally sketched by Y. Saitô.

earth, wholly consisting of the microscopic particles of plagioclase, a few fragments of pleochroic hypersthene, and little magnetite, but no glass splinters are seen. It reminds me of



Pro. 3.—Section exposed at the west coast of Jiri, Hoko.

the felspar sand that cover the flat and form the ground of Pampanga, north of Manila<sup>1)</sup>. After this short digression, I return to the former subject. Now, a yellowish-brown, loose sandy bed, 3 m. thick, comes below the middle flow, locally with limonitic nodules (Fig. 3). This is succeeded by another complex bed, 3 to 4 m. thick, made up of multifarious alternations of clays and sands, all retaining the original horizontal position. Then comes the third sheet of porous lava

of variable thickness, underlaid by a lignite bed, the last one can be only seen at low tide. The whole seems to me to be one complex bed belonging to later Teritary; and this profile serves as a type of the stratigraphical order of the island. After passing over the second 'haul-over' to the Fûkibi point (Plate V), opposite to Bakô, nothing but the two upper flows is exposed.

A table island, named Ko-sei-sho<sup>2</sup>, off the coast of Jiri, already referred to, is an erosion relie of the Basaltic mesa, surely connected in former times with the main island of Hôko. The adjoining wood-cut shows clearly the geological structure

B. Kotó, 'Geologie Structure of the Malayan Archipelago.' This Journal, Vol. XI., p. 113.

<sup>2)</sup> 虎井嶼.

and the general view, as seen from Jiri, exhibiting the two upper flows, mainly hidden by debris cones. This island served for the Chinese in former times for the strategic base against



F16. 4.—A view of the Isle of Ko-sei-sho, an erosion relic of Basaltic mesa, as seen from the coast of Jiri.

the Hollanders and Koku-sen-ya (Koxinga), in maintaining the sovereignty over her supposed vassal domain of Taiwan.

Starting again from Sei-shi-an, already referred to, and going round the south coast along the points of Kan-on-san'i and Kô-kaku<sup>2</sup>, Basaltic cliffs with underlying sandy bed, and sandy coves repeatedly occur as far as A-kan3. At Sa-kan4, a little south of the last-mentioned locality, fuller's earth similar to that of Bakô, is said to occur according to Tada and Ishii. Upon the walls of the cliff at the recesses of the coves are found, attached, according to Saitô, apparently recent shells, telling the fact that at no geologically remote period, probably Diluvial, a negative shifting of sea-level has taken place in this tract. We are, however, not informed of the height of the former level, as compared with the present; but at any rate it is of paramount importance for us to have been acquainted with this movement in view of the fact that on the opposite coast, i.e. on Front Taiwan, there are not wanting evidences tending to prove the negative change on the shore.

<sup>1)</sup> 觀音山 2) 候角 3) 鳥崁 4) 鑞管.

Between A-kan and Ri-sei-kaku<sup>n</sup>, the easternmost point of the island, a white sandy beach bounds the south shore.

All along the coast from Ri-sei-kaku to Hoku-rvo<sup>2</sup>, coral reefs limit the eastern shore, and the detritals derived from them form the beach-flat. It is a noteworthy fact that on the north side the coast is very deeply indented in the north-south direction, and the lowland, partly marshy, is covered likewise with coral sand. I may here mention an occurrence of coal which was once considered to be a very important natural resource of the island, though afterwards it turned out to be almost worthless and unworthy of public attention. At one of the points, called Kotô or dragon head, that stretches out northwards, a butte of Basalt, 22 m. high, elevates itself from the shore, and at its northern foot a seam of lignite, 5 feet thick, crops out with a sandy rock between the first and second flows, corresponding to the Arca zone in Gio-ô Island, already referred to. The exposure is meagre and soon disappears under the rubbish to be seen no more. This mineral combustible is but imperfectly incarbonized, and the woody structure is said to be yet well preserved.

From Sei-kei<sup>a</sup> through Kò-tei<sup>a</sup>, and Sha-kô<sup>a</sup> as far west as to the oft-mentioned Bakô, along the north coast, the two upper flows are the sole rocks that can be seen, being covered with an incoherent brownish, coarse and craggy earth.

#### HAKU-SHA ISLAND.

Haku-sha-tô,6 or the white sand island is bodily connected with Hôko through the intervening islet of Chû-don7, at the

<sup>1)</sup> 裏正角 2) 北紫 3) 西溪 4) 港底 5) 沙港 6) 白沙 7) 中墩。

two narrow neeks of the abraded second flow of Basalt, and forms a part of the geological unit, differing from them only in that here the interstratified sedimentaries seem to be wanting. The other features that strike the eyes of observers are firstly, the lowness of its relief, the highest point being Kô-don-san<sup>10</sup>, 36 m. high, and secondly, a considerable development of Alluvial accumulation of the shells and skeletons of low organisms, hence the name of the island. Cliffs, however, can be seen in its northern shore, exposing the youngest flow with its usual columnar structure at the water's edge. White sandy flats prevail throughout the rest of the lonely island, especially towards the Bay of Hôko, and the residual product of considerable thickness, derived from the Basaltic decomposition, covers the interior.

One thing worthy of mentioning is a sporadic occurrence of lapilli that had run aground on the east shore, probably from one of the Indonesian volcanoes. The pumiceous fragments, worn and rounded, belong to a Hypersthene-andesite with a highly pleochroic, rhombic augite, and this rock either massive or pumiceous can be seen in no other parts of the group.

The islets, Impai<sup>2)</sup> and Chô-sho<sup>3)</sup> or Bird Island, off the east coast, seem to be geologically identical, representing the erosion-relics of the Diluvial epoch. A luxuriant growth of coral reefs fringes the latter, as well as the neighbouring islets, just as in Haku-sha.

#### KIPPAI ISLAND.

Farther away in a northerly direction lies the islet of Kippai<sup>4</sup>, which is a low Basaltic flat, covered with half-

<sup>1)</sup> 後墩山 2) 員員 3) 烏嶼 4) 吉貝.

hardened for aminifer sand (Pl. II, Fig. 6.) of Recent age; fragments of corals, bivalves and serpula mixed with other components. The foraminiferal rock consists of millions of discoidal and spiral, water-worn shells. Rarely they have spines well-preserved. Viewing a section of the shell under the microscope, it is seen that the test consists of the tubulated proper walls of chambers, besides the canaliculated intermediate skeleton which forms spur-like marginal appendages, characteristic of Calcarina, and its external form and microscopic details agree well with C. Spengleri, Linné<sup>1)</sup>, dredged for the first time near the coast of Amboina at the depth of 1,425 fathoms. This species seems to be quite as abundant in the East Indian Archipelago, as we find here in the Pescadores. By wear and tear of rolling waves, the surface of the test becomes smooth, and the presence of spines can be usually only recognized in examining the structure of the supplementary skeleton which points to the former existence of some sort of prominence.

### GIO-Ô ISLAND.

Gio-ô, or Fisher Island, lies to the west of Hôko, and encloses with the latter the head-less Bay of Hôko, or rather an arm of sea. What has been said of other islands as regards the geology and the topography, holds true also of Gio-ô, with the differences, that the island is really table-shaped, bounded on all sides by cliffs, leaving no space for Alluvial deposits, excepting the shore and fringing reefs; and that the igneous sheet as well as the interbedded sedimentaries are developed to their full advantage, thus affording the best opportunities for geolo-

<sup>1)</sup> Challenger Report, 'Foraminifera.'

gists to get insight into the geological structure, and to study the stratigraphic details, of the whole Pescadores.

The oft-mentioned three flows and interstratified tuffites as well as the underlying bed are likewise present, and well seen

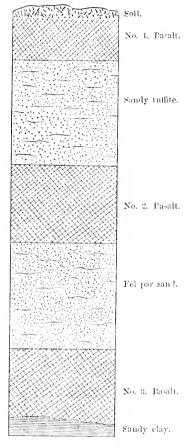


Fig. 5.—General profile as seen in the southern part of Gie-ô.

especially in that portion that lies southwards of Shô-chi-kaku.11 the Between last-mentioned locality that is situated in the middle of the island and Shûba-wan,2 good sections may be traced, as in fig. 5, in descending order. Under the superficial covering of the ferruginous soil decomposition from Basalt comes the No. 1. Basalt-flow, with its usual columnar structure, of about one foot, and sometimes disappears altogether. The third in the series consists of pelitic sand and loose sandstone, the latter being made up of muscovite, plagioclase, and Basalt-glass. Concentric nodules of hematite are frequentlv found in them. - Saitô is

fortunate enough to find in this complex bed casts of an Area and gasteropod (Turbo) in the matrix of ferruginous felspar sand with a little magnetite. Judging from the cast, the shell of the

<sup>1)</sup> 小池角 2) 葺馬彎.

Area is thick, egg-shaped, the ends of the margin obtuse-angled; the margin anteriorly rounded, posteriorly sloping; the beak prominent, anteriorly inclined, widely separated and inflated; coarse radial ribs more than 20 in number. Our specimen apparently resembles A. subcrenata, Lischke, though in details they may differ, if perfect samples are taken in comparison.

The next in the series is the porous, No. 2. sheet, underlaid by a fine felspar sand bed. Then the lowest, No. 3. sheet of 6-7 feet, often Agglomeratic; and lastly, the bluish-grey sandy clay, consisting of clay, muscovite, plagioclase and brownish opaque grains probably of Basaltic glass together with carbonaceous matter. It is remarkable that muscovite is more or less intermixed with in all the sedimentaries.

Before quitting Gio-ô, it should be remarked that the area north of Shô-chi-kaku, as well as the whole east coast is composed of the two upper flows only with or without interstratified beds; while the rest of the island, as may be seen in fig. 5, are built up of the second and third flows, accompanied with sedimentaries, unsurpassed in complexity and in thickness.

According to Tada, the islands of the Southern Group (Pl. IV.) of the Pescadores, are geologically of the same type. Counting southwards, they are:—Hattô, with the dependent isle of Shô-gun-ô; the Smaller and the Larger Biô-sho, so named eat islands from their appearance as seen from a distance; Tai-sho and Shô-hei with columnar Basalt; Tô-kitsu and Sei-kitsu, likewise Basaltic; all being encircled by coral reefs.

<sup>1)</sup> 八罩 2) 將軍嶼 3) 猫嶼 4) 大嶼 5) 小平 6) 東吉 7) 西吉·

#### III. Petrography of the Effusives.

The groundwork of the Pescadores is essentially built up of Basalts, making extensive flows to the water's edge, and the whole is encircled by the fringing reefs of corals, which, in parts raised above the water, connect many of the detached rocks with the shore, thus contributing greatly to the enlargement of the areas of the island. Each and every island visited by Tada and Saitô, presents the same physiognomy, and consists of the same black rock. The specimens, brought back from most of the islands, and of which descriptions will be given in the sequel, have a certain common feature which stamps them as genetically identical, and their field relations in different areas seem to point to a common centre of volcanic activity. They exhibit, however, a considerable variation of character. Thus from the same island, I have specimens at one place perfectly massive and compact, at another vesicular and porous, and sometimes Doleritic. Colours vary from black to bluish-grev in fresh ones, and through weathering the Doleritic and vesicular varieties become whitish or grey, while the compact rocks acquire a reddish brown tinge.

We are indebted to Mr. Y. Saitô, for characterising the different flows, and for tracing their vertical as well as horizontal distributions in the Northern group. According to him, there are three distinct Basaltic flows of nearly the same distribution, separated by long time-intervals which are represented in interbedded sedimentary rocks. Judging from the nearly perfect horizontality which the beds and flows keep in all the islands, it seems probable that there existed a lava field or volcanic mesa of considerable extent. But, on account of its remote age, pro-

bably later Tertiary, and of its insular position, waves gnawed the ground in time, finally reducing the once wide volcanic field into the ruins of islands, as we see at present. It is not easy to know the former extent, and the ancient surface feature, of this lava-flat; but, generally speaking, the relief becomes higher as we go southwards from one to the other in the islands of the Northern group. Saitô recognises, as I have already said, three lava-flows in the Northern group, viz., the uppermost or youngest being of columnar, the middle porous and vesicular, and the lowest also partly vesicular, and Agglomeratic. After the comparative study of the Basaltic rocks, to which the effusives exclusively belong, several important facts are brought to light, and now I am able to say, that the youngest flow (a, b and? d types) contains the iddingsitized olivine, at least in one type, and violet brown titan-augite; the second (c and? d types) the brown augite, olivine sometimes lacking, being often replaced by hypersthene; and the third (e type) the analcime-bearing. I will record first my observations on the component-minerals, and then give the special description of rocks.

#### A. Component-minerals of Basalts.

#### OLIVINE.

Olivine is rarely automorphic, but mostly xenomorphic, being the remains of resorption by magma. The olivine in the Basalts of the Hôko Islands seems to be of several varieties. Automorphic ones show vivid polarisation-colours, and alter usually into some red minerals. The xenomorphic type shows comparatively a low degree of polarisation, and suffers deep corrosion,

often being reduced to a mere grain, and is also traversed with fractural lines, from which the mineral begins to form a serpentinous substance. The olivines are undoubtedly the intratelluric products, being sometimes enclosed by an automorphic augite, and large individuals are habitually surrounded by heaps of the crystals of augite (in Andesites, instead of it often hypersthene). Inclusions of gas and liquid are not rare, and the octobedra of magnetite are also found in the olivines.

Zonal structure of olivine is, as is well known, of rare occurrence, and if it really exist, this could only be discerned either by measuring the optical angles at different portions, or by finding the altered zones in a crystal in consequence of the formation of the mineral rouge. The zone of the red mineral is not constant in position, for, it makes its appearance sometimes on the periphery, at other times in the interior; but, so far as my experience goes, the recurrent zones are never found. The condition under which the isomorphic shells of different chemical compounds are formed in the olivine, seems to depend, as Lagorio<sup>1)</sup> and Morozewics<sup>2)</sup> say, mainly on the Massenwirkung, that is, the degree of saturation of magma in certain temperature and pressure. In my slide, in which olivine has a red central zone (the Kippai Island specimen), magnetite is scarce, and large in its size and rod-shaped; while the magnetite-rich rock (the Hôko specimen) has an olivine with an external red zone. Here the magnetite occurs in small isometric crystals and grains.

The red mineral, that forms the periphery (Pl. I. Figs. 1, 4 and 5) and the kernel (Pl. I. Fig. 6), differs in habit. The

<sup>1) &#</sup>x27;Ueber die Natur der Glasbasis, sowie der Krystallisationsvorgänge im eruptiven Magma.' T. M. M. Bd. VIII, 1887.

<sup>2)</sup> Ibid. Bd. XVIII, 1898.

first has a facile cleavage but brittle, and consequently becomes lamellar like a brittle micaceous mineral. It is probably identical with the so-called biotite, which we occasionally find mentioned in petrological literature, as being formed from an alteration of olivine, just like as schillerspar has been considered to be a mica, as an alteration-product of enstatite. Recently, Iddings10 and Lawson<sup>2)</sup> described a similar mineral and the latter author named it iddingsite. In the second, we fail to find such a distinct cleavage, and it seems to me to be the same body which Michel-Lévy called the mineral rouge<sup>5</sup>. Now, a question suggests itself to me, whether the red micaceous mineral is identical with the mineral rouge or not? It is true that the former confines itself to the margin, and in the case where the entire substance of olivine has been transformed to this mineral, the process of alteration has started from the periphery, and it not infrequently happened to me to find every stage of progress from the very beginning to the complete alteration. The latter, on the contrary, starts from the centre in irregular patches, and gradually attacks the whole body but the clear and granulated, thin margin. formation of the red lamellæ begins with the development of a fine parting which appears like stripes, and which runs parallel to the vertical axis (Pl. I. Fig. 1); while cracks on the margin favour the olivine being changed into the red mineral in the centre.

In my opinion, there may be a slight difference in the

<sup>1)</sup> U. S. Geol, Surv., 'Monograph' XX., p. 388. Iddings identifies this mineral to thermophyllite, a foliated mineral having the composition of serpentine.

 <sup>&#</sup>x27;The Geology of Carmelo Bay.' Bulletin of the Department of Geology in the University of California, Vol. 1., p. 31. See also Pirsson's paper, Amer. Journ. Sci., XLV, 1893, p. 381.

<sup>3) &#</sup>x27;La Chaine des Puys et le Mont Dore,' Bull. Géol. Sor. France, 3me Serie, XVIII, 1890.

chemical composition of the two alteration-products, yet on the whole they must be practically identical. The lamellæ are oriented parallel to one of the pinacoids, as may be deduced from the position of the optic plane (in the fresh substance of olivine), which stands at right-angles to the easy cleavage (Pl. I. Figs. 1 and 5). Pleochroism is distinct; it is brownish-green in the direction of facile cleavage, but greenish-brown when at rightangles to it. Hence, c>a or b. Mügge<sup>1</sup>, however, says that the absorption is stronger in the direction perpendicular to the 'Längsrichtung' than in that parallel to it. Zirkel<sup>2)</sup> and Rosenbuch<sup>3)</sup> interpret the above statement in the terms, that the rays vibrating parallel to c absorb far less than those parallel to a and b. The observers, however, seem to have examined the mineral rouge. My observation, therefore, accords well with that made by Lawson for iddingsite; but it is not known to which pinacoid, 010 or 100, the lamellæ are parallel, though it is probable that the brachypinacoid is the lamellar plane, as may be inferred from the fact that the elasticity perpendicular to the lamellæ is greater  $(\mathfrak{A}=b)$  than that parallel to the c-axis, the latter corresponding to the mean axis of elasticity.

With HCl, the iddingsitic mineral becomes bleached, and then acquires a greenish-yellow colour, with corresponding decrease of pleochroism. Bearing in mind the fact of the brachypinacoidal lamellar cleavage, of the colour, and of the chemical composition which is a hydrous non-aluminous silicate of iron, lime, magnesia, and soda, I am rather inclined to consider the iddingsite to be a mineral approaching to basite. Prof. Rosenbusch<sup>41</sup>,

<sup>1)</sup> Newes Jahrbuch, 1883, H, S. 205.

<sup>2) &#</sup>x27;Petrographie,' Bd. I, 1893, S. 353.

<sup>3) &#</sup>x27;Physiographie,' Bd. I., 1892, S. 469.

<sup>4) &#</sup>x27;Physiographie,' 1892, Bd. I., S. 461.

in speaking of bastite, says 'die Umbildung scheint in hohem Grade durch die gleichzeitige Anwesenheit des Olivin und dessen Umwandlung zu Serpentine befördert zu werden.' Chemically speaking, there exists a close resemblance between iddingsite and the 'crystallised diallage' of Baste<sup>1</sup>, considering out of question a trace of alumina. Optical schemes differ, of course, in the two minerals, but I could not make out surely the optical orientation of iddingsite in my slides, on account of its extremely fine lamellar structure.

#### PLAGIOCLASE.

Plagioclase has, generally speaking, crystallised out in a single generation of the flow period. Differing from the Andesitic plagioclases which present various dimensions, the felspar of Basalt is uniform in size. It is, however, not wanting in large, phenocrystic crystals in some slides, which also belong to the products of the effusive period, slightly earlier in crystallisation than the ones in the general mass; for, the small laths of plagioclase are partly embraced by the phenocrysts,—a fact which also leads me suppose that the plagioclases have grown in a comparatively motionless magma. They show no signs of corrosion, so common in the olivine of the intratelluric origin, though the effects of tossing and fracturing of crystals are by no means seldom observed.

The *phenocrystic plagioclase* (*Pl. II, Fig.* 2) has a tabular form on M, somewhat elongated towards the vertical axis. Zonal structure is rare in contrast to the Andesitic felspar; the same

<sup>1)</sup> Hintze, 'Handbuch der Mineralogie,' Bd. II., S. 972.

is the case of glass-enclosures, save that ground-mass which fills up the rectangular space between the lamellae, showing as if the larger crystals have grown out by the apposition of numerous flowing lamellæ. Penck<sup>1)</sup> holds the same view, as given here. In consequence of this lamellar composition, which is one of the causes of the paucity of inclusions, both terminations of the ledges became indented and forked, after the manner of a parapet (Pl. I, Figs. 4 and 6., Pl. II, Fig. 5), a characteristic common to all the plagioclases of Basalts. It seems more reasonable to consider these monstrosities as incipient forms of growth, having simultaneously many centres of crystallisation in space. which in later stages have grown together to make up one individual with but internal complex compositions. Morphological and optical homogeneities are, however, frequently disturbed through the flowing motion and sudden cooling of the consolidating magma. Several stages of similar kind in crystallisation may be frequently observed under the microscope in the formation of artificial crystals.

Symmetrical but contrary extinction takes place at the maximum angle of  $33^{\circ}-35^{\circ}$ , with reference to the suture of the albite-twinning, and the extinction with regard to the pericline-lamellæ amounts to  $-16^{\circ}$ , showing that the plagioclase is of a basic labradorite. It is easily acted on by HCl. The Baveno twins were once observed.

The plagioclase in the ground-mass is lath-shaped, extremely slender, and polysynthetic; termination being also a parapet-like. The habit of crystals is prismatic, and such a form is said to be elongated parallel to the a-axis. This is indeed true; for,

<sup>1) &#</sup>x27;Studien ueber lockere vulcanische Auswürflinge.' Zeitschr. d. d. geol. Gesell. Bd. XXX., S. 101. Taf. V., Figs. 3, 5, and 7.

along the longest extension lies the axis of greatest elasticity, and there are tens of thousands of laths visible in microscope slides, with but a few tabular sections. Symmetrically opposite extinctions make the maximum angles of 23° to 25° with the suture of lamellæ. Microlithic sections twinned on the albite type extinguish at the angles from 0° to 26°, with reference to the longer dimension. According to Michel-Lévy, labradorite and albite have similar optical deportment, but as they do not usually come together, and as we are dealing now with a basic rock, the nature of the microlite should be considered to approach that of an acidic labradorite. In a few slides, the poles of the laths resolve themselves into a number of prisms, and such fine slender needles are scattered through the whole groundmass.

#### AUGITE.

Augite, so says Morozewicz<sup>1</sup> belongs to one of the 'rer-hangnissrollen minerals. It does not obey Fouqué and M.-Lévy's rule of crystallisation of silicates in the reversed order of fusibility, nor Rosenbusch's scheme of crystallisation according to acidity. It is rather subjected to the influence of masses, i.e. a degree of saturation under certain temperature and pressure. Under such circumstances, augite may form crystals before plagioclases, and at other times, just the reverse may occur, while in the third case they may individualise at the same time. According to the priority of secretion of either of the two minerals, in other words, the relative idiomorphism of one to

<sup>1) &#</sup>x27;Experimentelle Untersuchungen ueber die Bildung der Minerale im Magma.' Tschermak's Mitth., Bd. XVIII., S. 84.

another, various structures may be brought about under varying conditions, and these we find in fact in my slides.

As regards the forms of augite, it is sometimes idiomorphic, bounded by faces  $\propto P^{\infty}$ ,  $\propto P^{\infty}$ ,  $\propto P$ , and  $P^{\infty}$ , the first being well developed, consequently the crystals become tabular; at other times granular, needle-shaped, in ophitic plate, and in partial crystals. Prevailing colour is either violet or yellowish-brown. It is to be expressedly remarked that the typical Basaltic augite with a tinge of violet occurs only in the Pescadores, and in the dykes of Basalt near Taihoku and Taikokan, in Formosa. My long experience forced me to conclude that, in Japan proper, the Basalt with the violet augite is confined to the northern Kiûshiû, and Chiu-goku, in Hondô, as far east as the provincial boundary of Tajima and Tamba. The same type of Basalt is also known to be wide-spread in Korea, Liau-tung, and Mongolia. Thus the distribution of the Basalt with the violet titaniferous augite marks a definite area, being, so far as my knowledge goes, confined to the inner side of the festoon islands and the adjoining continent in Eastern Asia, constituting the well-defined Japan-China petrographical province. Larger crystals show a zonal structure, coloured intensely on the periphery, and the hourglass structure occurs frequently with deeply-coloured, additive cones in the prismatic zones, which have at the same time a greater angle of extinction. Pleochroism is stronger in the direction parallel to the c-axis. Polarization-colours are generally weak in comparison to those of the Andesitic augite. Twins on  $\propto P^{\frac{1}{2}}$  have a suture, running just along the middle of the body of the crystals. Crystals often form stellar aggregates; they are generally free from foreign interpositions, excepting the larger ones which have sometimes enclosures of glass and magnetite.

#### HYPERSTHENE.

Hypersthene takes the place of olivine in some Basalts of the Pescadores; consequently the presence of one totally excludes that of the other,—a state of thing quite exceptional to the modern Japanese Andesite of a glassy, black, porphyritic type, in which both minerals appear always concomitantly. We have then the Hypersthene-Basalt, in lieu of the Basalt proper. It is a noteworthy fact that this stray variety of rock seems to be widespread, at least in my specimens, in the out of the way islets, such as Impai, Kin-sho, and Hattô, the only exception being the one from Sei-kei (West Valley) in Hôko, though I could not find a sufficient reason accounting for the special distribution of this hypersthene-bearing rock.

It is usually a comparatively easy task to discriminate hypersthene from olivine, but in the present case some difficulty is experienced in making out for certain the presence of the former.

In regard to the form, the (1) hypersthene is extremely slender, being about six times longer than broad, and, as being of the intratelluric origin, it has a marginal zone deeply corroded and partly granulated, and has indefinite faces at the poles of the crystals (Pl. II, Fig. 3). I observed once a morphotropic growth of a highly-polarising, monoclinic pyroxene around a hypersthene, just as is the case in Andesites. Cleavage is developed along the longest extension of the crystals. In a patch of a coarse aggregate which appears as an endogeneous or homogeneous enclosure in the finer general mass, the (2) hypersthene comes together with plagioclase and augite, and in this case the hypersthene occurs in broad plates (Pl. II, Fig. 4), with only a few

traces of cleavage, but with numerous fissures; and has an appearance exactly like olivine.

The hypersthene possesses a brown colour, and its pleochroism is scarcely discernible. In favourable cases, the ray vibrating parallel to the c-axis is slightly green. Sections present a rough surface, owing to its having a high index, approaching to that of olivine; its polarisation-colour is grey.

From the brief diagnosis, given above, of the hypersthene, its cleavage, colour, non-pleochroism or very weak if present, high index, but low magnitude of refraction, extinction-direction, and similar chemical composition,—these several physical properties afford no means of discriminating it from a fresh olivine. Olivine has, however, a lighter colour, and has usually but one trace of cleavage in a section. The hypersthene on the other hand possesses the characteristic traces of prismatic cleavage, which in a random section gives scarcely a clue to distinguish it conoscopically from monoclinic pyroxene. A basal section, once observed, presented a square outline, truncated little at the four corners.

From the combined evidence of more slender section, of the want of decomposition-products, of indifferent behaviour towards common acids, of the presence of comparatively numerous traces of cleavage, I infer, in the Basalts, the presence of a hypersthene. It is to be remembered that the prismatic sections of olivine show also a low colour of polarisation, exactly like that of a hypersthene. It seems to me that the hypersthene in the Hôko Basalts stands in its chemical composition near to that of bronzite. The want of a distinct pleochroism may be attributed to the same cause. Axial angles, therefore, become large, and the axial poles were not observed in any of the pinacoids by ordinary methods.

#### APATITE.

Apatite occurs in the Doleritic or Anamesitic rocks in the form of extremely fine needles, devoid of terminal faces, being colourless, and always traversed by transversal fissures. Its crystals sink almost to a minimum size, and are not, comparatively speaking, so large as those found in the typical European Dolerites; and for this reason they might be easily mistaken for the microlites of felspar which often resolves from the poles of a larger crystals in Basalts. The apatite is typically found in the three slides only, which are in my possesion (Kippai and Hôko), and both are magnetite (not ilmenite)bearing rocks. The crystals are dark-margined, owing to the total reflection of light on the prismatic faces; and sometimes a single brown-coloured axis entirely or partially runs through the crystal. A grey or light-brown variety, so often found in Andesites, is entirely absent, though a dark-brown crystal of an apatite-like mineral was once observed with strong absorption parallel to the prismatic axis. The sure criterion of the presence of apatite can only be found in its hexagonal cross-section.

#### ANALCIME AND NATROLITE.

A cave-rock in the southern Gio-ô, presents an anomalous habit; a slide made of it contains a colourless mineral in angular or polygonal interspaces between the crystals of plagioclase (Pl. II, Fig. 5). It shows no signs of any crystallographic face, nor cleavage, but only has a frittered appearance, being traversed with irregular cracks, and also being pierced through in all directions with the needles of apatite which is excessively rich

in this rock. The polysomatic mineral has a smaller index of refraction, when compared with that of the accompanying plagioclase, as may be easily experimented upon by Becke's method. These colourless patches, as a rule, behave optically isotropic; at times, however, faintly double-refractive, and separate into several optical fields. They readily dissolve in HCl, with the formation of the cubes of rock-salt. The same patches frequently resolve themselves into a radial-fibrous, somewhat brownish and highly double-refractive body (well seen at the margin in the lower, left quadrant in Pl. II, Fig. 5) with the positive sign along the axis of the needles.

The polygonal base-like mineral, moulded upon plagioclase and augite, seems to be identical with what Büking<sup>1)</sup> calls the 'Basis zweiter Art,' and is allied to the pitchstone-glass of Hunter and Rosenbusch.<sup>2)</sup> Recently, this base was studied with great zeal by the American petrologists, Lindgren,<sup>3)</sup> T. F. Williams,<sup>4)</sup> Kemp,<sup>5)</sup> Fairbanks,<sup>6)</sup> Cross,<sup>7)</sup> Coleman<sup>8)</sup> and Pirsson<sup>9)</sup>; the last author especially paid close attention to this subject, in making careful analyses and also recalculating the analytical result, obtained by Hunter. From his study, Pirrson is forced to the conclusion that the so-called colourless base has exactly the

<sup>1) &#</sup>x27;Basaltische Gesteine, etc.,' Jahrb. K. K. preuss. geol. Landesanstalt. 1880, S. 153, und 1881, S. 606.

<sup>2) &#</sup>x27;Ueber Monchiquite, ein camptonitisches Ganggestein aus der Gefolgenschaft der Eleolithsyenite,' Tschermark's Min. Mitth. XI, 1890, S. 445.

<sup>3)</sup> Proc. Cal. Acad. Sci., Vol. III, 1390.

<sup>4)</sup> Cited in Pirsson's paper.

<sup>5) &#</sup>x27;Trap Dikes,' Bull. 107, U.S. G.S. 1893.

<sup>6) &#</sup>x27;On Analeite Diabase from San Luis Obispo County, California,' Bull. Geol. Depart. Univ. Cal., Vol. I. p. 27?.

<sup>7) &#</sup>x27;An Analcite-Basalt from Colorado,' Journ. Geol. Vol. V. p. 684.

<sup>8) &#</sup>x27;A new Analcite Rock from Lake Superior' Journ. Geol. Vol. VII, 1899, p. 482.

<sup>9) &#</sup>x27;The Mochiquites or Analcite Group of Igneous Rocks,' Journ. Geol., Vol. IV. 1896, p. 679.

same chemical composition as that of *analcime*, and the physical properties observed give no hinderance to the assumption that this component actually is that mineral. He thinks the analcime is primary, having been formed from the magma, containing water and much soda, under pressure with considerable rapidity.

From what has been stated before, I have also, to all appearances, the primary analcime in the interspaces of the components in the Basalt from Gio-ô, and the radiating bundles of a strongly birefringent natrolite are formed secondarily from the analcime through a molecular rearrangement. Both components make their appearance with the dodecahedric networks (Pl. II, Fig. 5) of the skeleton magnetite which occupies the other portion of the slides.

#### THE IRON ORES.

Both ilmenite and magnetite are present, and they usually belong to a single generation, and indeed the product of the effusive period, as the iron ores were not found enclosed in the olivine of the intratelluric crystallisation. Both ores, especially the ilmentite, have crystallised *later* than plagioclase, but slightly prior to, or contemporaneous with, the monoclinic pyroxene. The ilmenite and magnetite are, under the microscope, not easy to be distinguished, as every petrographer will agree, if crystal forms are not serviceable for their diagnosis.

The *ilmenite* is, however, tabular and needle-shaped in section in the Basalt with a strong lustre and a violet tinge, when seen by reflected light, on the flanks corresponding to the thickness of slide. The laths are slender, appearing merely as lines, and cross several crystals of felspar and augite, mean-

while the substance of the ilmenite entirely disappears when traversing other crystals, and comes again into view in the same direction as a continuation of the interrupted crystals. Unfortunately basal sections were not frequently observed, and this was the greal obstacle in ascertaining the presence of ilmenite n microscopic analysis. The ore with above-mentioned lamellar habit occurs exclusively in a coarse-crystalline type of intersertal, or ophitic structure, irrespective of hypersthene or olivine-bearing Basalt; and this fact lends evidently a strong support to the view advanced by K. Hofmann, 1) that the ilmenite accumulates in the lower portion of lava-flows, and in that which has crystallised under high pressure, while the magnetite is rich in the upper part that has consolidated under a low pressure. Fr. Sandberger<sup>2)</sup> says also that Basalts may be classified into Dolerite and Basalt proper, by the presence of ilmenite in the former and magnetite in the latter. These fruitful ideas inaugurated by both authors, now unfortunately passing into oblivion, deserve the careful attention of petrologists.

A slide of the Basalt from the islet of Hattô was treated for a considerable length of time with a strong hydrochloric acid without any appreciable result. A large quantity of the pulverised sample of the same specimen was then digested in boiling HCl with the addition of tin-foil, and the solution was coloured slightly violet, showing the presence of titanium in the dissolved portion of the ore. Ilmenite also occurs, according to Vénukoft<sup>(3)</sup>, very abundantly in the Basalts of Mongolia, and even transparent lamellæ were found by him, just as in the Pescadores rocks. The ilmenite is fresh and leucoxene not noticed.

<sup>1) &#</sup>x27;Basalt von Pakony,' Zeitschr. d. d. geol. Ges., XXIX., 1877, S. 191.

<sup>2)</sup> Rosenbusch, 'Mikroskopische Physiographie,' II., 3te Auflage, S. 1015.

<sup>3) &#</sup>x27;Les Roches Basaltiques de la Mongolie,' Bull. Soc. belge de Géologie, etc. T. II., p. 443.

In my few slides of Basalts, bearing the *iddingsitised* olivine, ilmenite seems to be wanting, though the rocks approach to a Doleritic type, being replaced by magnetite. I cannot say positively that this rule holds true for all the iddingsite-bearing Basalts.

The magnetite is, on the other hand, the prevailing ore in the compact Basalt, and in the Limburgitic type, in the form of isometric crystals and dust, occurring either in the general mass, or else enclosed in augite and olivine. The face of the crystals shows a metallic lustre with a tinge of blue by reflected light. The dust is sometimes peripherally altered into a blood-red iron-glance. A slide made of a chip from Hôko, was digested in HCl with the addition of KI; and then the black ore, therein contained, was entirely removed, and the solution not coloured when tested with tin-foil, proving thus the presence of a pure magnetite. As it is already stated above, the magnetite-rich, compact type seems to make up the upper portion of the thick flows of the Hôko Basalts.

In the Anamesitic type from the islet of Gio-ô, we find beautiful networks of the skeleton-crystals of magnetite in a devitrified mesostasis within the polygonal spaces between crystals. They are the dodecahedric dendrites, consequently the skeletons intersect each other at the angles of 60° and 120°, and are said to consist of garnetohedrons. They all go into solution by treating with HCl. Morozewics<sup>1)</sup> tells us that the spire and filigree-work of the skeleton magnetite, crystallising out of the magma rich in iron oxides, consist of minute octahedra, arranged rectilinearly in the direction of the crystallographic axes with secondary and

<sup>1) &#</sup>x27;Experimentelle Untersuchungen über die Bildung der Minerale im Magma,' Tschermak's Mittheilungen, 18, 1898, S. 90.

tertiary offshoots. This mode of growth, the octahedric dendrite, so called by Morozewics, is well known in petrographical literature, since the publication of Prof. Zirkel's<sup>1)</sup> work. On the other hand, it is said that the dodecahedric dendrite, as is in the present case, is formed out of the magma poor in iron-oxides.

#### B. Special Description of Individual Occurrences of Basalts.

#### A. THE GRANULAR TYPE.

(Pl. I, Figs. 1 and 2.)

As seen by the naked eye, it is greyish-black and compact, with the dots of olivine which is the only visible component of the whole mass. This type is represented by two specimens from Hôko, and one from Hakusha.<sup>2</sup> Microscopically it is holocrystalline with the smaller phenocryst of olivine, imbedded in the still finer aggregate of the ground-mass.

The fineness of the ground-mass, however, varies in different specimens, and even in the same slide. Some portion of the same slide is, therefore, extremely rich in idiomorphic augite to the total exclusion of felspar and olivine, but with small patches of brown glass. Were this portion independently developed, it would be fitly called the *Augitite* (Fig. 2). It is the local assemblage of augite within the rock, and that mineral es-

<sup>1) &#</sup>x27;Die mikroskopische Beschaffenheit der Mineralien und Gesteine.' Leipzig, 1873, S. 244.

<sup>2)</sup> Collected at Ryô-l-ô-sun (蒙望山); and, according to Mr. Saitô, it appears in I. horizon, i. e., the uppermost sheet, consequently the youngest of all the lavas of the Hôko Group (Pt. I, fig. 1).

pecially accumulates near the margin of the secretionary mass, the augite being sometimes arranged along the linear common base, with the free ends of the crystals toward the interior. These phenomena indicate that the lava had consolidated in a quiet state.

The relative proportion of augite and plagioclase is also various, and in the cases where the former outweighs the latter, the olivine increases in its quantity and comes also in the ground-mass, as a product of the crystallization of the effusive period; and at the same time the texture of the rock becomes finer. If, on the other hand, the plagioclase becomes predominant over the augite, then, the texture gets coarser and more crystalline, and the distinction between phenocrysts and ground-mass is not then commonly well marked. Apatite and ilmenite seem to occur in the latter variety only, the ilmenite is sometimes transparent with a deep brown colour.

The only mineral that serves as the phenocryst is olivine. Its forms are various, owing to the various degrees of resorption. Most have partial crystallographic faces with deep indentations of corrosion, and a drop-like black iron-ore and felspars were formed in those spaces. Sometimes the act of corrosion has advanced so far that there remain but patches as the relics of a large crystal, and the eating away of the body by the magmatic menstruum proceeds always from the lateral pinacoids. As usual, the crystals of the olivine are not fresh; but the routine of change is the same in all. They become fibrous and lamellar, parallel to one of the lateral pinacoids, the altered portion being yellow or brown, according to the degrees of transformation. The mode of change is similar to iddingsitization (Figs. 1 and 2).

The ground-mass consists, first of all, of the crystals and

grains of augite, all of a *violet*-brown colour, besides the grains of olivine, and the laths of the multiple-twinned plagioclase, the octahedra and dust of magnetite, and ilmenite. The texture of the rock is crystalline and typically *granulitic*. In a coarse variety, the idiomorphic augite with hour-glass structure forms stellar aggregates, and these aggregates closely resemble the glomeroporphyritic phenocryst.

#### B. THE TYPE OF THE IDDINGSITE-BEARING BASALT.

Megascopically this type is greyish-black Anamesitic-looking, and finely uniform-granular, owing to the nearly equal size and form of the laths of plagioclase which predominates quantitatively over the other components.

The characteristic features of this group are firstly, the presence of large phenocrysts of olivine which is more or less iddingsitized; secondly, the majority of the augite is xenomorphic or granular, and of small size, and these grains are grouped together intersertally with the devitrified glass between the laths of plagioclase. The structure is typically intersertal. The prominent characters distinguish this group from the rest of the Basalts. This type is represented in my slides from the Pescadores by three specimens, one from Kippai, and two from Hôko, one of which was struck off at the locality Tai-san; according to Y. Saitô, it forms the uppermost flow there. The same may be said of the specimen from Kippai, since the youngest

<sup>1)</sup> 字天山, 譯霍鄉, 澎湖. The rock efferve ces with acid. The microscope discloses the fact that the radially arranged fibres of calcite fill up the polygonal spaces between other components, showing bars which correspond to the position of crossed nicols.

lava-flow is the only effusive that can be met with on that island.

The olivine is the sole phenocryst; it is variable in size (the largest one measures even 5 mm.), irregular in distribution, and multifarious in form, some having partial crystallographic faces, while others have none of them. The iddingsitization is peculiarly inherent in the olivine of this rock-group, and I refer the readers for further details to the topic: "component-minerals" p. 18 et seq. By the way, I have only to mention that the name iddingsite may conveniently be applied to a special transitional form of the alteration of an olivine which, after passing this stage, changes into dirty-green spherulitic fibres of an optically positive character.

In the felspar-rich rocks (Pl. I, Fig. 6), which are prevalent in the group under question, the plagioclases are all approximately of the same size, and surpass the augite both in dimension and quantity; while in the augite-rich rocks (Pl. I, fig. 4), the plagioclases are of two generations, and the larger ones behave porphyritically towards the minor ones. They are lath-shaped, and multiple-twinned, the terminations being imperfect and sheafy, and these laths are thrown together in an orderless plexus, which eminently characterises the structure of normal Basalt in contradistinction to that of Andesite.

The augite is all of a single generation, consequently uniform, but inferior in size to the plagioclase and olivine. Some are rudely idiomorphic, but by far the most of it is granular, occurring in groups, and filling the angular spaces left by the laths of plagioclase. The angite is, as usual, of a violet-brown colour, but in the specimen from Tai-san, it is almost colourless in sections. It is free from foreign inclusions, and

the hour-glass structure is faintly indicated in some individuals. In the coarse, felspar-rich specimen, the *iron-ore* is present only in small quantity (*Pl. I, Fig.* 6), but comparatively large, lamellar and flat with glittering bluish lustre on the perfect cleavage-surface. It looks rather more like ilmenite than magnetite. Stiff, slender *apatite*-needles, sometimes with a brown canal traversing the whole length, are particularly abundant, being scattered through the whole mass.

In the dark fine specimens (Pl. I, Figs. 4 and 5), small regular crystals of magnetite are plentiful, and in these slides, I found abundantly the small laths of twinned plagioclase, which resolve at the ends into slightly diverging columns (Pl. I, Fig. 5), and these may be easily mistaken for those of apatite, if needles are found detached from the waist. Optical properties are not independtly shown in them, on account of their extreme thinness. Similar bodies are noticed by H. S. Washington in the sanidine of some Ischian Trachytes and named by him keraunoid. He and also Lehmann<sup>2)</sup> attribute the splittings and ramifications from the main crystal to the existence of internal tensions in felspar, but the cause of the existence of such tensions remains to be solved.

The glass together with the augite fill up the polysynthetic space left by the laths of plagioclase. The glass-base is coloured bottle-green, sometimes dirty brown, and devitrified in various ways. It consists of polarizing scaly aggregates of vermiform, spherulitic, or, irregular shapes. Sometimes fascicular and radiating needles, which are colourless and birefringent, are imbedded, in the green base as a product of devitrification. The

<sup>1) &#</sup>x27;On some Ischian Trachyte,' Journ. Amer. Sci., May, 1896, p. 380.

<sup>2) &#</sup>x27;Molecularphysik,' I, 1888, S. 378.

needles may possibly of a felspathic nature and such a structure is termed *variolitic* by Harker, though in the present case those circular, whitish spots, called varioles, are wanting. Green, fresh base is here and there also found between the angular spaces.

Thin lamellæ of rugged outlines, with violet-brown colour, may be frequently noticed in all of my slides, and they closely resemble those found as interpositions in a hypersthene. I conjecture the mineralogic nature of these plates to be ilmenite.

#### C. THE OPHITIC TYPE.

(Pl. 11, Fig. 2)

This type is represented by a single specimen from Hôko, and Gio-ô² respectively, and two from Haku-sha, though the 'lie' is not known to me exactly; but it is highly probable that samples are taken from the *second* sheet which is separated usually from the uppermost columnar flow by an ash bed of a certain thickness. It is a greyish-black, Anamesitic rock, with the *brownish*, lath-shaped phenocrysts of plagioclase (4 mm. length). This is the coarsest type of the Hôko Basalts, and is the one rich in plagioclase in comparison with ferro-magnesian silicates; it seems to have solidified in the lower portion of the lava flow.

Under the microscope, it shows a porphyritic, hypocrystalline, diabasic structure (Fig. 2) with the ophitic plates of augite of considerable dimensions, enclosing the laths of plagioclase which lie in all possible directions. The augite is of a

The aggregates of felspar-microlites or fibres with fan-like or sheaf-like groupings.
 They may be closely packed to make up the entire mass of a portion of the rock (Basalt)."
 Petrology for Students.' 2nd. Edit., p. 191 and 201, Fig. 41 A.

<sup>2)</sup> The exact locality being Sho-chi-kaku, (小池角) at the middle of the island.

kind of light-brownish colour, and its plates are often multipletwinned, and enclose, besides plagioclase, a number of round and corroded crystals of olivine which is for the most part changed into green, pleochroic fibres; the iddingsitization of the olivine was so far not observed. The plagioclases are of two generations (Fig. 2), the larger, probably intratelluric, species has fissures (see Fig. 2) filled with films of brown hydrous sesquixide of iron, which cause the phenocrystic feldspar to appear macroscopically like an olivine. The plagioclase is partially embraced by the ophitic plate, while the smaller laths became entirely enclosed in it. The polygonal interspaces, when not occupied by augite, are otherwise filled up with the fibrous devitrified glass, the latter containing globulites, sometimes dendritic, and apatite; and the thick lamellæ of ilmenite traverse the base, but not the plate of augite, consequently the crystallisation of the ore must have taken place posterior to that of the pyroxene. Sometimes the greenish-yellow augite is coarse-granular, and in this case the structure approaches to that of intersertal. Magnetite seems to be wanting. Owing to the coarseness of the structure, the rocks are often porous, and the polygonal, angular spaces are often filled up with banded, purplish chalcedony.

#### D. THE TYPE OF THE OLIVINE-LESS BASALTS.

(Pl. I, Fig. 3; Pl. II, Figs. 2 and 3.)

The olivine-less, hypersthene-bearing Basalts are represented in my collection by two specimens from Wampai<sup>11</sup>, and one from each of the following islands, Hôko<sup>21</sup>, Kin-sho<sup>31</sup>, and Hattô-sho<sup>41</sup>. They are megascopically wet-grey, and fine-granular,

<sup>1)</sup> 灣貝 2) Sei-kei 西溪 in Hôko 3) 金嶼 4) 八罩島.

the general microscopic aspect being a crystalline Andesite-like. They are all extremely rich in augite, and the structure is granulitic. The felspars are of two generations (Pl. II, Fig. 2 and 3), and the rock is consequently porphyritic, owing to the presence of a few large crystals of plagioclase, though this structure could not be easily recognised as such in the present group.

The phenocrystic plagioclase is narrow-tabular with a few twinning lamellæ (see Fig. 3), and is remarkable in its being traversed through by sets of cracks which run approximately parallel with each other. In one instance, only one lamella, out of many twinned parts after the pericline law, is provided with closely set fissures. This anomalous feature can be seen in all the specimens of the present type, but not common in others, and the same peculiarity recurs also in augite whose granular aspect is due in great measure to the same cause. I cannot offer at present a satisfactory explanation to account for this phenomenon; but, as Judd savs, it might in part have been caused by a slow but constant movement of a crystallizing magma, and also chilled suddenly, perhaps by the access of water at the final stage of consolidation. I may here adduce in support of my ground a fact of the special distribution of the Hypersthene-basalt which, so far as I am aquainted with, occurs only in the outlying islets, excepting the locality Sei-kei, on the north coast of Hôko, which also lies not very far from the present sea-shore.

Hyperthene occurs exclusively, though insignificant in quantity, in the form of phenocryst (Pl. II, Figs. 2 and 3) and takes the place of olivine in the present rock-group. It is sedge-like in general shape, and granular in its margin (especially in Fig. 2), being fringed with grains of common augite, whose presence becomes strikingly apparent between crossed nicols,

on account of their vivid colours of polarisation in contrast with the grey tint of the hypersthene in the interior. Pleochroism is scarcely perceptible. Traces of a few rough cleavages run through the hypersthene lengthwise, and as in the case of the plagioclase, it is trasversed with many fissures. The hypersthene is of intratelluric origin, and has the general aspect of its having been worn out caustically and frittered, and the peripheral accumulation of augite, already referred to, seems to have some genetic relation with the act of degeneration.

Large, monoclinic *augite* sometimes makes its appearance in company with the hypersthene and plugioclase, forming local patches of secretional origin, with the *hyperitie* structure.

The ground-mass, which constitutes the main bulk of the rock, consists of laths of plagioclase and grains of the frittered and corroded augite, together with rugged clumps of magnetite. The relation of the first two components cannot be told in a few words. In one instance, the mutual relation is such that we could almost say it is ophitic; in another, it is intersertal in company with a little remnant of brown glass, while in the third, no such arrangement could be discovered, but a simple aggregate of felspar and grains of augite, thereby calling forth the structure which is termed granulitic. The augites of both generations are of yellowish brown and not violet-brown.

Shingly tridymite fills up polygonal spaces, and the loose brushes or tufts of either plagioclase or apatite are thrown through the whole mass. A doubtful iddingsite (Pl. I, Fig. 3) was once observed, and some rocks are calcareous too. The stratigraphic position of this type is not known to me. It may be the lava of either the first or the second flow.

# E. THE TYPE OF THE ANALCIME-BASALTS. (Pl. II, Fig. 5.)

This to the naked eye is macroscopically deep-grey, and fine-granular. Under the microscope it is hypocrystalline and more or less porphyritic, either the xenomorphic olivine or the aggregate of the automorphic angite being the phenocryst, or sometimes both. The texture varies within a wide range, but generally speaking is coarse (Fig. 5). The porphyritic elements, however, differ generally not much in size from the crystals of the ground-mass, and the mode of arrangement of the several components is granulitic. Plagioclase predominates over augite in quantity; and magnetite is not plentiful, and completely soluble in HCl. The paucity of iron-ore causes the rock to appear of a grey shade.

Olivine occurs as a phenocryst in the xenomorphic grains, a few of which have been reduced even to mere flecks through gradual Cleavages are not noticeable in contrast to other resorption. olivines, but in stead of them there are curvilinear cracks, conforming approximately in their direction to the boundary of The substance of the olivine is colourless, and usually resorption. more or less altered into a greenish or yellowish, fibrous substance (not iddingsitic). Brown decomposition is quite foreign to the olivine of this type. The present olivine seems to belong to a variety rich in magnesia. Phenocrystic pyroxene is scantily present in some, but none in others. The augite is of the typical Basaltic variety, with a violet-brown type, possessing the hourglass structure, and idiomorphic, flattened on the orthopinacid. It occurs singly or in stellar aggregate. There is no felsparphenocryst.

The ground-mass consists of laths of plagioclase, crystals

and crystalloids of violet-brown augite, magnetite, and xenomorphic olivine, with the interstitial mass of analcime and The laths are multiple-twinned with the parapet-like terminations (Pl. II, Fig. 5) produced by the shifting of lamellæ to the one end or the other with reference to the adjacent plate. The slide treated with HCl shows a considerable corrosion of the interior lamellæ of the laths, while the exterior remains intact and fresh, as if a frame is enclosing the hollow space. The crystals of a violet-brown augite of the short prismatic habit, rather flattened towards the ortho-axis, are freely developed, or occur in clusters. The augite and plagioclase must have, therefore, crystallized simultaneously, and at their contact the one is partially penetrating the other and vice versa. Magnetite is idiomorphic, but frequently possesses irregular outlines, owing to the penetration of the crystals of plagioclase, augite, and apatite, and the larger crystals are anhedrons, as they are moulded upon the neighbouring laths of the plagioclase. The magnetite is comparatively large and few, excepting its dendritic skeleton crystals which are found abundantly in the specimen from Gio-ô, in company with devitrified glass. In the specimen, which is wanting in dendritic magnetite, there are brown, biotite-like lamellæ usually in association with the hexahedral iron-ore. The lamellæ are anisotropic, and distinctly pleochroic, and the mineral is conjectured to be ilmenite.

It is of no small interest to note the presence of analcime. It occurs sporadically rather in large patches in the cuneiform spaces left by other crystals. It is generally fresh and colourless, and isotropic, but often shows the optical anomalies so common to this mineral. At times, the analcime resolves into a dirty, fibrous natrolite (as in the left, lower margin in Fig. 5). The

analcime seems, so far as my experience goes, to be exclusively confined to this type, though it is possible that the colourless base in minute interspaces of other Basalts of the Hôko Group, might turn out to be that mineral, if the means are at hand in ascertaining its presence.

Another accessory to be mentioned is apatite in colourless prisms, which is especially plentiful in this type.

The colourless base and analcime are rather unexpected quests in the basic, black rock, such as we have here, and the mode of occurrence is that they fill up the polygonal interspaces left by the crystals of other components of the rock. If we accept the primary origin of the analcime, as Pirsson<sup>1)</sup> would do, it is all the more very striking to see that the residuum of a Basaltic magma should have an exact composition of Na<sub>2</sub>O·Al<sub>2</sub>O<sub>3</sub>·4 SiO<sub>2</sub>·2H<sub>2</sub>O. Yet the analcime seems to all appearances to be of primary origin, if we take into account the perfectly fresh state of the rock in which it is found, and not only in the Basalts of the Hôko Group, but in the Teschenite of the Nemuro promontory in Hokkaidô, I had several occasions to observe the same mode of occurrence of the analcime, so that it could not be attributable to a mere accidental circumstance to find it in such state, as several foreign writers also noticed the same. It excludes the idea of its having replaced the base which formerly occupied the place of the now-existing analcime.

The present mode of occurrence of the analcime may perhaps be explained by supposing that, when the Basalt was consolidating on the surface in a quiet state, carrying in it the intratelluric olivine, the newly created crystals, such as those of playioclase, augite,

<sup>1) &#</sup>x27;The Monchiquite or Analcite Group of Igneous Rocks.' Journ. Geol., Vol. IV., 1896, p. 679.

magnetite, apatite, together with the olivine had then sunk down, and formed the heap of crystals at the bottom, meanwhile the unconsolidated residuum of the magma was actually slowly flowing through the sieves of crystal-heap, or changed its chemical composition through diffusion, after the manner of liquation as in a metallurgical process. And, then, the solution having the composition of the hydrous alumino-sodium-silicate has finally crystallised out in the interspaces of the meshes of crystals. Similar process can be frequently observed during the formation of crystals on the stage of the microscope. If this be the actual condition under which the Analcime-Basalts have consolidated, considerable leaching and percolation must have taken place during the formation of rocks, and the structure of such a rock should better be called the 'leached.' This structure is therefore properly seen only either in the granitic or in the granulitic rock, consequently it is wanting in the family which has a fluxional arrangement of the components.

The Analcime-Basalts are represented in my collection by three specimens from Gio-ô, and one from Hôko. The hand-specimen from Nai-an<sup>1)</sup> in Gio-ô, is, according to Y. Saitô, said to occur at the water's edge, the main portion of the flow usually lies under the level of sea, and constitutes the third sheet of lavas, and is the lowest, consequently the oldest of the accessible lavaflows of Gio-ô.

Other Analcime-Basalts of the Hôko Group no doubt belong to the same horizon.

<sup>1)</sup> Nai-an 內安, 漁翁島.

### THE ISLAND OF KÔTÔ', (BOTEL-TOBAGO).

Starting from Makian<sup>2</sup>, one of the Spice Islands, a long chain of the Molucean volcanic system runs upwards, and joins at the solfatarie volcano of Api, in Mindanao, with that of the Sangirs, that comes from the north end of Celebes. The united system of volcanoes in the Philippines, then, receives the name of the Mayon system. It goes right through the whole breadth of Mindanao, and enters Caminguin, Leyte, Biliran, and, afterwards, the peninsula of Camarines of south Luzon. It is in the last-named region that the volcanic activity of the Philippines is fully displayed. Albay or Mayon stands foremost in rank among the mighty cones. For a while, we lose sight of the chain northwards under the Pacific bottom, and it reappears in full force at the crater of Cagua near Cape Engano, in north Luzon.

The northern prolongation of the Mayon system may still be traced through the little-known Babuyans,<sup>5)</sup> the Batans, and the Bashi islands. All are said to be of volcanic origin. Among the Bashi or Vasshi<sup>5</sup>, the five larger islands, going from the south to the north, are Liayan, Mabudis, Tanem, Maysanga, and Tami, the last being the largest of the forlorn isles. An active volcano is said to exist in the southern region (?), spreading fire and destruction.

The Balintang Canal at 20° N. lat. separates the Japanese

<sup>1)</sup> 紅頭.

<sup>2)</sup> B. Kotô, 'On the Geologic Structure of the Malayan Archipelago,' This Journal, vol. XI, pages 111 and 118. Wichmann calls the chain the 'North Molucean bow,' Der Wawani auf Amboina und seine angeblichen Ausbrüche, III.' Tijdselev, v. h. Kon. Nederl. Gen., Jaurgang 1899, S. 32. This bow is now said to start from Batjan, lying to the south of Makjan, loc. cit. S. 14.

<sup>3)</sup> Kotô, loc, cit. p. 118.

<sup>4)</sup> The Japan Mail, August 10th 1897, 'Forlorn isles.'

domain from that of the United States. Within the Japanese waters lie the Batans, the Bash islands, the rocks of Gadd and Forest Belle, the islands of Shô-Kôtô (Little Botel-Tobago) and Kôtô (Botel-Tobago), and, lastly, Kashô (Samasana), as the continuation of, I conjecture, the Mayon system of volcanoes (Fig. 1).

The smaller isle of Kôtô is, geologically speaking, entirely unknown, but the Larger Kôtô has been several times visited by the Japanese, since the first landing of a staff of the governorship of Taiwan, in April, 1897. Among our University men, Mr. Tada stayed there a week collecting zoological specimens, and, lately, Mr. Torii remained longer in this lonely island among the aborigines for his anthropological study. I myself have not had the opportunity of visiting it, though the island has been within my sight for a week long, while travelling the pathless beaches of south-eastern Taiwan.

The island of the Larger Kôtô (Fig. 1) lies in a south-eastern direction about 50 miles off the coast of Pinan, and 35 miles north of north-east from the Cape of Galambi in Taiwan. Its north-south extent is 3 ri and the breadth  $1\frac{1}{2}$  ri, with the circumference of 9 ri. It is the abode of 1,500 nude aborigines. Seen from a distance, this scapula-shaped island appears plateaulike in general profile, crowned by a prominence of 120 m., somewhat excentrically situated in the north; and is bounded by steep declivity all round the coast, so that it leaves only a narrow patch of lowland on the south-western shore, which serves at the same time for the chief anchor-ground of this islet.

Being situated amidst the stormy and swift *Kuro-shiwo* current, the narrow beach is highly cobbly, as may be seen from Mr. Torii's photographs: and the steep cliff undoubtedly owes its present form to the abrading action of dashing waves.

Fringing reefs are said to skirt the shore, some portion attaining double the man's height above the water's edge, indicative of a recent negative shift of the relative levels. It seems to me probable, that they are not the reefs of Neocene time, which usually attain a considerable height of more than 200 m., as in the Apes Hill of Takao, but those of a comparatively recent date, possibly representing a Diluvial formation. The plateau-like elevation, which faces the sea in cliffs, seems in parts at least in the north-east point to consist of volcanic agglomerate. A greater part of the interior seems to be built of volcanic rocks with a gabbro-like plutonic mass as the foundation of the island exposed at the west coast, but their mutual relations and area of distribution are quite unknown to me.

In the following, I will give a succinct account of rocks, kindly placed at my disposal by Messrs. Ishii and Torii.

#### A. FELSPAR-BASALTS.

(Pl. III, Figs. 3 and 4.)

My slides of Basalts and Andesites are prepared from chips of water worn gravels, used as weights attached to a fishing net of the aborigines.

The Basalt is rather porous, greyish-brown mass with a few phenocrysts of a brown olivine (1-2 mm.) and black common augite. Under the microscope, the olivine occurs in two generations (Fig. 4). Its forms are acute six-sided, sometimes nearly square, truncated at corners, but mostly corroded and disfigured, with a few traces of basal cleavage. The crystals are slightly decomposed in their margin, being either yellow

or brown; but as a whole the interior is fresh. It is the iron-rich variety—hyalosiderite, as is proved by the micro-chemical reactions, which show only a trace of magnesia. The olivine encloses a large quantity of regular octahedra or elongated crystals of the brown, transparent picotite, mixed with the crystals and dust of magnetite.

Plagioclase, as a phenocryst, is observed only once in my three slides; it is long-rectangular in form, with negative crystals, filled with a gas. The crystal is multiple-twinned, extinguishing light symmetrically with the maximum angle of 32°; consequently it is the calcium-labradorite. The augite is rather automorphic, showing, however, a slight corrosion marginally. This character is common to all of the specimens. The crystal occurs in polysynthetic twins; the colour yellowish-green and non-pleochroic. As usual, it has glass-enclosures with air-pores. Sometimes, the augite is internally and nucleally resorbed, leaving an accumulation of grains of the same in its place. The augite is of nearly the same size as the olivine.

The ground-mass is seen, under the microscope, to make the main bulk of the rock. The micro-phenocrysts of olivine and augite are the same in habit as the macro-phenocrysts. The augite is in a few cases fringed with skeleton-crystals. They are inbedded in the plexus of the felspar-laths and clumps of magnetite, rudely showing a flow structure. The laths are twinned simply or polysynthetically, and in many case hollow, with the very thin external rim, partially or entirely filled up with glass. So far as I am aware<sup>1</sup>, such skeleton-crystals of felspar seem to

<sup>1)</sup> The same skeleton laths are observed by E. Elich in the Amphibole-pyroxene Andesite from the Rio Blanco, West Cordillera, Ecuador. Reiss u. Stübel, 'Reisen in Süd-Amerika, Das Hochgebirge der Republik Ecuador, I.; Petrographische Untersuchungen, I. West Cordillere,' S. 163.

be of extreme rarity. The laths extinguish light symmetrically but in the contrary direction at an angle of 26°-27°, proving the felspar to be more acidic than its phenocryst. Interstitial space is occupied by a brown glass which contains globulitic and rod-shaped bodies. From the foregoing descriptions, it is evident that the Basalt of the Island of Kôtô does not properly belong to the category of normal Basalt with violet augite, presenting the intersertal structure, which is so common in the rocks of the Hôko group, already described. Here exclusively monoclinic augite presents the character of diopside. Both the olivine and augite, all being equally corroded, present so great a variation in size from the macroscopic to the microscopic dimensions that I could not discriminate the products of the intratelluric from the effusive period of consolidation. The ground-mass, as I have said, is highly felspathic, and the structure is Andesitic and hypocrystalline-porphyritic, somewhat resembling a pilotaxitic type. Richness in olivine and paucity in iron ores, as well as globulitically granulated mesostasis make the rock approach to a navitic structure (Fig. 4), the only difference being the presence of feldpar-laths in the ground-mass. The rock seems to me to be a lava-flow, consolidated rapidly, accompanied by a brisk liberation of gas from the cooling magma.

#### B. HORNBLENDE-ANDESITES.

(Pl. III, Fig. 5.)

I have three specimens of rocks in Torii's collection, belonging to the same category. They differ in colour consequent on the various stages of decomposition. A fresh one is greyish and porous, speckled with phenocrysts of hornblende (2 mm. in length).

Plagioclase is long-rectangular along the zonal axis at rightangles to 010, and tabular when parallel to that face (Fig. 5). It varies in size so that between the phenocrystic felspar and that of the ground-mass we could find a series of dimensions. Zonary structure is typically developed in almost every individual, especially on the tabular section of 010. It contains, as usual, glass arranged in zones; sometimes encloses crystals of augite and hornblende, parallel to base and the positive dome; it extinguishes light in symmetrically opposite directions with the maximum angle of 30°-34°. The extinction observed on 010 amounts to 15° with reference to P/M, the trace of the pericline twins making 1.5° with P/M on the same face. These rough observations all point to the labradorite-nature of the felspar. Hornblende occurs only as the phenocryst and small in quantity. It is a brownish-green variety of optically normal character. The crystals are all corroded and enclosed by the opacitic margin (Fig. 5) which is composed of confused aggregate of crystalloids and grains of monoclinic pyroxene, and clumps of magnetite. The pyroxene appears in tolerably large size that it could be optically ascertained. Sometimes the substance of the margin has been replaced by brownish, double-refractive fibres. In one slide the body of the hornblende is impregnated with countless swarms of black dots which lend to the crystal a darker shade. With high powers, they resolve into glass-enclosures with bubbles.

Augite occurs sporadically as a phenocryst. Its coarse distinct cleavage, pale colour, and small angle of extinction (less than 32°) prominently characterise this pyroxene, and contrast pronouncedly with the brown, Andesite augite. That it is diopside is highly probable, but not proven. In one slide, I observed porphyritic aggregate of needles, producing the glomeroporphy-

ritic structure, and they look more like a druse than like a mass of crystals, having a mutual relation, characteristic of plutonic rocks. Thus our augite is remarkable in many respects.

The ground-mass consists mainly of the idiomorphic plagioclase, long-rectangular or square in shape, and of various sizes, with some degree of parallel disposition. The square sections of the microliths show occasionally truncation of corners by domal face and at other times slightly diverge from rectangularity on the edge 001:010. The traces of cleavage run parallel to the same edge, and the sutures of twins run vertically. Symmetrical extinction takes place at 30°-32° with reference to the same trace, showing that the plagioclase stands just at the middle of the series between the sodium and the calcium labradorite1. According to Becker, these square sections, which are prismatic sections in vertical positions, are very convenient for the determination of the microlithic plagioclase. Intermixed with the felspar, we find the less idiomorphic crystals of pale augite, together with rounded magnetite and the crystals of apatite. The cuneiform space left by minerals being filled with the brown glass, densely charged with transparent augite. The structure of the rock is therefore that kind which we call the 'orthophyric.' In the  $\beta$  variety, minute felspar-needles make the greater part of the ground-mass, exhibiting the typical pilotaxitic structure.

#### C. APOANDESITES.

## (Pl. III, Figs. 1 and 2.)

One variety is whitish, bleached and compact, the other is green through the presence of a chloritic mineral, having a por-

<sup>1)</sup> Becker, Amer. jour. Sci., May, 1898.

phyritic structure with the phenocrysts of plagioclase and horn-blende. They are much speckled with glittering iron-pyrites (a large black spot in Fig. 1), which likely attracted the attention of Mr. Narita, who had brought back the specimens to Tai-hoku.

The phenocrystic plagioclase has a tabular form being nearly equidimensional. It has a distinct zonary banding, like the preceding rock. Contrary symmetrical extinction of about 33° on both sides of the trace of the albite twins shows the plagioclase to be a labradorite of a similar composition as in the rocks, just described. Hornblende is entirely decomposed (in the right halves of Figs. 1 and 2) into an aggregate of pistacite, chlorite, and calcite-films, which together form the pseudomorph after the hornblende of a prismatic habit with the combination of 010, as may be conjectured from the original outlines of the now altered mass. The chlorite possesses the normal character, and pleochroic, showing a green shade parallel to the axis of fibres, which corresponds to  $\mathfrak{A}$ . The epidote occurs in tufts and in rugged plates.

The ground-mass consists of very fine laths, simply twinned, and they are arranged in more or less parallel disposition around the phenocrystic felspar. These minute crystals of felspar swim within the chlorite-lamellæ, mixed with the felspar-microlite, magnetite and the pyrites, the last does not contain any trace of copper. This Apoandesite is no doubt derived from the  $\beta$  variety of the Hornblende-Andesite, already described, by the pneumatolytic process which caused the impregnation of the pyrites in the rock-mass.

#### D. AMPHIBOLIZED GABBRO.

A dark-greyish, coarse rock of gabbroitic aspect, in which a cleavable hornblende lies after the manner of plutonics, and a plagioclase is moulded upon the amphibole. Patches of epidote and iron-pyrites complete the list of megascopic elements. Under the microscope, the greenish-brown hornblende is for the greater part altered into a nearly isotropic lamellæ of chlorite, calcite-films, and common epidote. The hornblende has been so highly altered that the original substance remains but in few stripes. The plagioclase-anhedra possess only a few twinned lamellæ, besides the Carlsbad type of twins. Suitable section could not be found for ascertaining the nature of the plagioclase. The general deep-greyish appearance of the felspar is due to the presence of a pennine-like chlorite in the fissure of it. Common epidote occupies the place of the felspar and hornblende in rugged plate. Crystalloid of apatite, full of air-pores, was only once observed.

I conjecture this rock to be a *metagabbro*, though a diallagelike augite was never seen in my slides. This gabbroitic mass probably makes the foundation of the island, and crops out on the *west* coast, together with the Apoandesite and Serpentine.

#### E. SERPENTINE.

Associated with the above rock, there occurs a Serpentine which is yellowish-blue in its general appearance. Under the microscope, the whole mass presents between crossed nicols a beautiful lattice-work, which is a characteristic feature of its having being derived from an amphibole. There are found intermixed with the Serpentine a little quantity of iron-ore.

### THE ISLE OF KASHÔ (SAMASANA).

(Pl. III, Fig. 6.)

Kashô is a forest-covered, conical volcanic island (Fig. 1), only 8 km. in circumference, skirted by fringing reefs. The inhabitants are of the mixed blood of the Chinese and the Malays. According to Mr. Ishii, who gave me a rock-specimen, the island is Andesitic, consisting of Pumice and lava-flows, and carries two craters. My slide shows the rock to be the Hypersthene-hornblende-Andesite.

To the naked eye the rock resembles very closely those of Héradaké, in Shinano, and Hakusan in the Kaga province. is greyish-looking, with the only phenocryst of hornblende, measuring 5 mm. by 2. The hornblende is the largest of phenocrysts (on the right half of Fig. 5), broad-columnar in form in combination of 110 and 010, and has always thick margin of opacite. The hornblende has dark-brown colour, and optically normal. It encloses the grains of felspar after the fashion of poikilitic plate, especially on periphery. This fact conclusively shows the simultaneous crystallisation of the hornblende in its later period with the forerunner of plagioclase. The formation of these crystals might have taken place at the close of intratelluric period of the magma. The opacitic margin consists, as usual, of the grains of monoclinic pyroxene and magnetite. They seem to have been formed by resorption and re-combination through the gradual caustic action of the surrounding magma upon the already existing hornblende, at a slightly lower pressure and in the upper column of effusive lava than the situation in which the original amphibole has crystallized out. The majority of crystals seems to have been eaten up by

the magma, so that there remains nothing but the accumulation of magnetite-dust in the place of the hornblende.

A brown pleochroic hypersthene occurs in few quantity, and small in size and less idiomorphic when compared with the amphibole. Its base shows no axial poles, but symmetrical hyperbolas; it forms penetrating twins upon a domal face, and often moulded upon plagioclase. The plagioclase is of tabular or long-rectangular shape; extinguishes symmetrically in the direction at about 30° against the trace of the albite-twins, and the trace of pericline lamellæ makes -5° to -10° on 010 with P/M, indicating the presence of labradorite. The albite-lamallæ are clear and definite, but the width varies much from one lamella to another, and even in the same the width varies from one point to another,—these are also said to characterise labradorite. Zonary banding is pronounced, the interior abounds in glass-enclosures, with the clear shell of different optical orientations. We meet often with the broken crystals, from which it may be inferred that the rock is a lava-flow. The ground-mass consists of a plexus of augite-needles in a colourless base, intermixed with a somewhat larger plagioclase of a tabular, or longrectangular form, after the manner of a micro-phenocryst. Twinned slender sections show symmetrically the opposite extinction at an angle of 20°, indicating that the felspar in the groundmass is andesine in lieu of the larger, phenocrystic labradorite. Magnetite abounds in the glassy base. Tridymite fills free spaces in imbricated scales.



# CONTENTS.

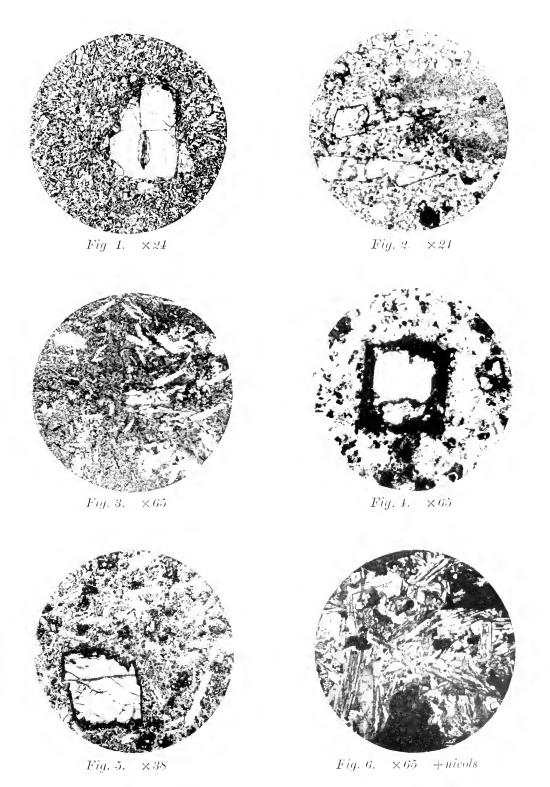
								Ρ.	AGE:
The Hôko Group (Pescadores)	•••							•••	1
I. Introductory								•••	1
II. Stratigraphical Characteristics		•••				,			õ
Hôko Island									5
General outlines of the geo	ology	of	the is	land			• • •		5
Local details of geology									8
$Haku$ -sha $Is$ /and $\dots$ $\dots$									12
Kippai Island									13
Gio-ô Island						• • •			14
III. Petrography of the Effusives									17
A. Component-minerals of Basa	lts							•••	18
Olivine									18
Plagioclase									22
Augite									24
Hypersthene								•••	26
Analcime and Natrolite									28
The Iron Ores									30
B. Special Description of Indiv	ridua	1 0	curr	ence	of B	asalt	s	• • •	33
a. The granual type	• • •	•••					• • •	•••	33
b. The type of the Iddings	site-b	earii	ng B	salt:	· · · ·			•••	35
c. The ophitic type									38
d. The type of the Olivine	e-less	Bas	salts				• • •		39
e. The type of the Analcit								• • •	42
The Island of Kôtô (Botel-Toba	go)	•••						•••	<b>4</b> 6
a. Felspar-Basalts									48
b. Hornblende-Andesites									50
c. Apoandesites					•••				52
d. Amphibolized Gabbro		•••							54
e. Serpentine					•••	•••	•••	•••	54
The Isle of Kashô (Samasana)									55

# PLATE I.

(PHOTOGRAMS.)

#### PLATE I.

- Fig. 1.—A fine compact basalt, with comparatively large phenocryst of olivine which is more or less iddingsitized. The ground-mass consists of small crystals and grains of augites, granular olivine and the laths of plagioclase, with the structure typically granulitic. Bô-ryo-san, Haku-sha Island. P. 33.
- Fig. 2.—The same rock-type as the preceding, but rather coarse. On the right side in the figure is a augititic patch, composed of exclusively the crystals of augite in the base. Hôko Island, P. 33.
- Fig. 3.—Olivine-less basalt from Hattô, Southern Group, and it probably belongs to the same type as Figs. 3 and 4 in Plate II. A doubtful olivine is present in the form of chloritic patches, but no visible hypersthene. General mass consists of a plexus of fine grains of augite and fine laths of plagioclase in the base. This is quite an anomalous rock. P. 39.
- Fig. 4.—Iddingsite-bearing basalt with a large idiomorphic olivine, externally changing into iddingsite. Magnified 65 diameters. Hôko Island. P. 35.
- Fig. 5.—Rock belonging to the same type as the preceding. It is also from Hôko Island. Olivine on the left side of the figure shows various stages of iddingsitization.
- Fig. 6.—Also iddingsite-bearing basalt, with olivines changing from the interior, as may be seen on the lower side of the figure. Magnified 38 diameters and not 65, as is stated in the Plate. Nicols crossed. Kippai Island.



imp. Tokyo Printing (a., Ltd.

4		

# PLATE II.

(PHOTOGRAMS.)

### PLATE II.

Fig. 1.—Iddingsite-bearing andesite, magnified 65 diameters, showing the typical intersertal structure. Olivine is here changed internally into a red mineral, which the writer believes to be iddingsite, as is well seen on the lower right octant in the figure (pp. 19 and 35). Kippai Island.

Fig. 2.—The slide of ophitic basalt (p. 38). Shô-chi-kaku, the Island of Hôko.

Fig. 3.—Olivine-less hypersthene-bearing basalt, with two large crystals of hyperthene in the centre of the figure. The structure is granulitic. The Isle of Wam-pai. P. 39.

Fig. 4.—The same rock-type as the preceding, but with intersertal structure. Local patches of hypersthene, augite and plagioclase, with the hyperitic structure. Sci-kei, the Island of Hôko. Pp. 39 and 41.

Fig. 5.—Analcime-basalt from Nai-an, Gio-ô. It has granulitie structure. White patches are filled with analcime, and a dirty portion at the middle of the field is the secondary natrolite. P. 42.

Fig. 6.—Foraminiferal rock, consisting of discoidal and spiral, water-worn shells of *Calcarina Spengleri*, besides fragments of corals, bivalves and scrpula. In natural size. Kippai Island. P. 13.



Fig. 1.  $\times 65$ 



Fig. 2. ×24



Fig. 3.  $\times 24$ 

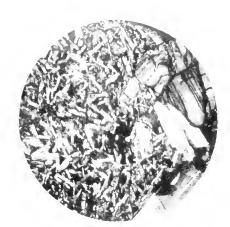


Fig. 1.  $\times 38 + nicols$ 

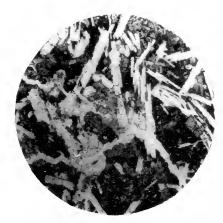


Fig. 5.  $\times 65$ 

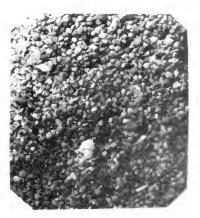


Fig. 6 Nat. size



### PLATE III.

(PHOTOGRAMS.)

#### PLATE III.

The Plate III illustrates the type-rocks from the Isles of Kôtô (Botel-Tobago), and Kashô (Samasana).

Fig. 1.—Apo or altered andesite, magnified 65 diameters, showing a phenocrystic hornblende, with opacite margin (on the right side of the figure). The hornblende is entirely decomposed into an aggregate of pistacite, chlorite, and calcite-films. Plagio-clase is much decomposed. A dark spot (in the lower left octant) is the iron-pyrite (p. 52).

Fig. 2.—The same slide under crossed nicols.

Fig. 3.—A porous, greyish-brown basalt, with a rather large corroded olivine (on the left of the figure). The ground-mass, which encloses a corroded diopside-like angite, is highly felspathic. The structure is hypocrystalline-porphyritic, approaching to the pilotaxitic type (p. 48).

Fig. 4.—Another basalt, with abundant olivine of various dimensions. It contains globulitically granulated mesostasis, and the structure is navitic (p. 50).

Fig. 5.—Hornblende-andesite, with dark hornblende-crystals, surrounded by opacitic margin (on the upper and the lower end of the figure). The structure is orthophyric (p. 50).

Figs. 1-5 are all from the rocks of Kôtô.

Fig. 6.—Hypersthene-hornblende-andesite from the Isle of Kashô, with a large phenocryst of hornblende (on the right half of the figure). It is enclosed by a thick margin of opacite, but enclosing the grains of plagioclase after the fashion of poikilitic plate (p. 55).

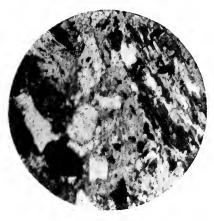


Fig. 1.  $\times 65$ 

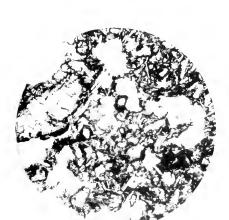


Fig. 3.  $\times 38$ 

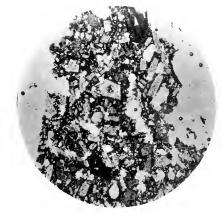


Fig. 5  $\times 38$ 



Fig. 2.  $\times 38 + nicols$ 

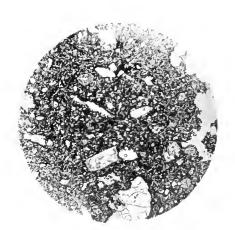


Fig. 4.  $\times 38$ 

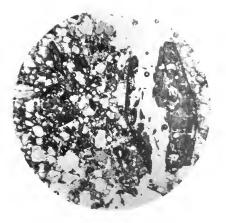


Fig. 6.  $\times 38$ 

		•

# PLATE IV.

(MAP.)

### PLATE IV.

The Plate IV shows the bathometric condition of the neighbouring seas of the Pescadores or Hôko Group. It seems to me that the North Group forms itself an independent centre of extravasation of magma, in contrast to the South or Rover Group, from which the Northern is separated by the incurve of the forty fathom-line,—the position indicated by the Rover Channel. Both groups are, however, located at the north-eastern end of the Formosa Bank, which is disconnected on the east from Taiwan by a channel of the same name (p. 3).

THE HÔKO GROUP.

Kotô, The Hôko Group, etc. Jour. Sc. Coll. Vol. XIII. Pl. IV. 25'lat. Wilat. Soliver Strain 3 Chû don North Group 5 23°30'lat Ku-sho Bin-sho South Group Scikichi To-sho her Tôkichi Formosa Bank 23'101. H9 E. long. 30-40 5 = 40 Fathous 20-30

Scale 1:680,000.

# PLATE V.

(GEOLOGICAL MAP.)

### PLATE I.

The Plate V is intended to show the geographical distribution of the Tertiary basalts with intercalated sedimentaries and several Recent formations, one among the latter being the coral-reefs which fringe the coast all round. The topographic basis for the geological map is compiled by myself from various sources, the data being supplied chiefly by Mr. Y. Saitô, who also offered me assistance in colouring the geologic elements represented on the map.

MAP OF THE HÔKO GROUP. Kotô. The Hôko Group, etc. Pore & Bannet



# Change of Volume and of Length in Iron, Steel, and Nickel Ovoids by Magnetization.

Ву

H. Nagaoka, Rigakuhakushi,Professor of Applied Mathematics.

AND

**K. Honda,** Rigakushi, Post-graduate in Physics.

With Plates VI. & VII.

1. In our former paper, we described some effects of magnetization on the dimensions of nickel and iron, as well as those of hydrostatic pressure and longitudinal pull on the magnetization. We then showed that there is a reciprocal relation between the two, and that the Villari effect in iron is a natural consequence of the observed changes of dimensions. Unfortunately on that occasion the range of the magnetizing field was limited to a few hundred C.G.S. units, so that the investigation of the behaviour of these metals in high fields was reserved for further experiments. In addition to this, the ferromagnetics were not of a shape to be uniformly magnetized with the exception of the iron ovoids. It was therefore thought desirable to repeat

Nagaoka and Honda, Journal of the College of Science, 9, 353, 1898; Phil. Mag. 46, 262, 1898.

the experiment on ovoids of ferromagnetic metals, and so to extend the investigation into still stronger fields.

- 2. In his well-known researches on the changes of dimensions of iron and other metals by magnetization, Bidwell<sup>1)</sup> pushed the field strength to 1500; in the present experiment, the field strength is greater than that of Bidwell by 700. In addition to ordinary soft iron and steel ovoids, wolfram steel from Bohler in Vienna was tested with a result which showed a remarkable difference from ordinary steel as regards the change of dimensions wrought by magnetization. As was generally supposed, the change of volume is very small in iron and nickel in weak fields, but with strong magnetizing force the effect becomes generally pronounced.
- 3. The apparatus already described was used in measuring the change of length and of volume. A small alteration was made in the arrangement of the magnetizing coil. Owing to the strong magnetizing current, special arrangements were made for keeping the interior of the coil at a constant temperature. A double walled tube of brass was inserted in the coil, and a constant stream of cold water was passed in the interspace for more than an hour before each experiment. As the resistance of the coil was only 0.562, the rise of temperature was so small, that the ferromagnetics placed in its core were scarcely affected. The change of length was measured by an optical lever, as before described. For measuring the change of volume, the ovoid was sealed in a glass tube with a capillary neck (internal diameter about 0.4 mm.) and so placed in the tube

<sup>1)</sup> Bidwell, Phil. Trans. 179, 205, 1889.

Nagaoka, Phil. Mag. [5] 37, 131, 1894; Wied. Ann. 53, 487, 1894.; Nagaoka and Honda loc. cit.

that it rested in the axial line, and never came in contact with the wall of the tube. The magnetizing coil and the tube were placed in a horizontal position. The motion of the meniscus was measured by a microscope provided with a micrometer ocular. For more minutely detailed particulars, we must refer the reader to the former paper.

3. The following are the dimensions of ovoids used in the present experiments:

Specimen No.	Metal	a (em.)	c (cin.)	v (c.em.)	ρ	N
1	Nickel	0.750	12.50	31.50	8.86	0.125
2	"	0.500	10.00	10.48	8.86	0.0848
3	Soft iron	0.750	12.50	31.45	7.84	0.125
4	,,	0.500	10.00	10.53	7.83	0.0848
5	Ordinary steel	0.750	12.50	31.60	7.83	0.125
6	,,	0.500	10.00	10.57	7.81	0.0848
7	Wolfram steel	0.750	12.50	31.82	7.90	0.125
8	"	0.500	10.00	10.53	7.95	0.0848

a gives the semi-minor axis, c the semi-major axis, v the volume,  $\rho$  the density, and N the demagnetizing factor of the ovoids. The volume of each specimen was measured by weighing the ovoids in water.

The elastic constants of the metals were measured by flexure and torsion experiments on rectangular prisms made from the same specimens as the ovoids. The prisms were 14.6 cm. long and 0.896 cm. square in cross-section.

Metal	E (C.G.S.)	K (C.G.S.)	$\theta$
Nickel	$2.07 \times 10^{12}$	$0.771 \times 10^{12}$	1.082
Soft iron	$2.10\times10^{12}$	$0.800 \times 10^{12}$	0.844
Steel	$2.04\times10^{12}$	$0.838 \times 10^{12}$	0.384
Wolfram steel	$2.02\times10^{12}$	$0.849 \times 10^{12}$	0.306

E gives Young's modulus, K = n. Thomson and Tait) the modulus of rigidity, and  $\theta$  a constant defined by the equation

$$\frac{E}{2}\left(\frac{1+2\theta}{1+3\theta}\right) = K.$$

The magnetization of each of these ferromagnetics was determined by the magnetometric method, after the ovoids had been carefully annealed, with the following results:

Nick	el (2)	-	Soft ire	n (4)	Steel	(6)	Wolfram	steel (8)
H	1		Н	I	Н	1	H	I
0.7	24.2	,	1.0	62	1.9	23	2.7	18
1.4	49.8		$2.5^{-1}$	160	2.4	44	6.8	65
3.0	138.6		4.3	291	6.9	183	12.6	193
5.4	238.0		9.5	587	9.7	279	20.2	498
10.9	336.8		12.7	750	13.1	385	25.8	748
37.8	395.7		9.61	948	23.3	651	44.5	992
74.1	420.0		37.2	1111	32.3	815	83.6	1116
125,3	434.5		99.6	1255	50.2	984	118.0	1170
174.6	438.7		155.5	1309	116.3	1196	191.0	1224
240.3	440.7		270.3 +	1400	174.4	1260	344.6	1301
481.4	443.4		$433.6^{\circ}$	1479	345.0	1379	512.3	1348
674.2	444.5		584.6	1520	520.2	1440	666.6	1373
914.0	446.8		792.8	1546	873.7	1489	940.3	1400
1233.0	447.7		992.6	1562	1149.8	1512	1213.3	1423
1747.0	448.7		1585.8	1607	1822.6	1549	1674.9	1452

#### Change of Length.

- 4. Iron (Fig. 1).—The change of length experienced by soft iron is too well-known to need any description. The ovoid elongates in weak fields till it attains a maximum, being longer by about 3- to 4-millionths of its initial length; it then decreases in length and becomes shorter than in the unmagnetized state. The contraction goes on gradually increasing, and, in the present experiment, it does not seem to reach an asymptotic value, even in fields of 2200 C.G.S. units, where the contraction amounts to about  $\frac{1}{100,000}$ . The present result agrees qualitatively with Bidwell's experiment, but the contraction is much greater. The discrepancy is perhaps to be chiefly accounted for by the difference of shape.
- 5. Steel (Fig. 1).—Ordinary steel behaves just like iron, the difference being the smallness of elongation and contraction, while the field at which the elongation vanishes lies in the stronger. The field of maximum elongation in wolfram steel is greater than in ordinary steel or iron, that of no-elongation in the unannealed state being several times greater than in iron or ordinary steel. Such a field lies in H=1200. When the wolfram steel is annealed, the retraction after reaching the maximum takes place very slowly and the characteristic as regards the field of no elongation becomes exceedingly pronounced. From the curve of length change, it does not appear that it will ever cut the line of no-elongation even in intense fields.
- 6. The curve of elongation (in dots) plotted against the intensity of magnetization is given in Fig. 1. The change of length

at first takes place very slowly, but on reaching saturation, the rate of decrease becomes very rapid. So far as the present experiment goes, the rate does not diminish except in annealed wolfram steel, in which we notice a slight flattening.

7. Nickel (Fig. 1).—The behaviour of annealed nickel ovoid as regards the length change is nearly the same as that already observed by one of us. With an increasing field, the contraction reaches an asymptotic value, which in the present case is greater than that obtained by Bidwell from experiments on a nickel wire. The explanation of this discrepancy is to be sought for partly in the difference of shape, and partly in the difference of treatment, as will be clearly illustrated by experiments on the change of volume. We have also reason to believe that repeated annealing alters the elastic behaviour of ferromagnetics as regards the strain wrought by magnetization. Plotting the curve of length change against the intensity of magnetization, we find a slight bend when the magnetization becomes saturated and the contraction approaches its asymptotic value.

### Change of Volume.

8. Experiments by several physicists prove that magnetization produces change of volume in ferromagnetics, in contradiction to the popular belief which is based on Joule's experiment. The alteration of volume accompanying the magnetization of ferromagnetics is generally very small in weak fields, but as will be seen from the present experiment, the phenomenon becomes more marked as the field is made stronger. As we have already remarked, the change of volume as measured by Cantone<sup>1)</sup> in an iron ovoid must

<sup>1)</sup> Cantone, Mem. della R. Accad. dei Lincei 6, 487, 1891.

have been exceedingly minute as the magnetizing field was very small. Dr. Knott<sup>1)</sup> has published several papers on the change of internal volume of ferromagnetic tubes, showing that iron, nickel, and cobalt are subject to the change by magnetization. As our former result regarding the same question was somewhat different, especially in the case of nickel, we have thought it advisable to settle the discrepancy by fresh experiments.

- 9. Iron and Steel (Fig. 2).—Preliminary experiments on soft iron and steel ovoid showed that considerable increase in the volume change takes place as the ovoids are annealed. The increase becomes more significant as the field is made stronger. In steel, the effect of annealing is greater than in iron. In strong fields, the volume change of the annealed steel ovoid is nearly twice as great as in the unannealed state. Wolfram steel is very little affected by annealing as regards the volume change, but the change itself is much greater than in nickel or iron. The motion of the capillary meniscus in the dilatometer can be easily followed by the naked eye. The curves in Fig. 2 have been plotted from measurements made on annealed ovoids.
- 10 Nickel (Fig. 2).—As specimens of nickel almost always contain traces of iron, the change of volume will probably depend on the chemical nature. In addition to this, the mechanical process which the metal had to undergo before it could be brought to a form suitable for experiment, must have substantially altered its elastic behaviour.

The nickel rod, which we used in the former experiment, was hammered from a nickel plate to a prism of square cross-section. It contained 1.75 % of iron, besides traces of manganese and earbon. The ovoids used in the present experiment

<sup>1)</sup> Knott, Trans. Roy. Soc. Edinb. 38, 527, 1896; 39, 457, 1898.

were prepared from a thick plate, and were nearly pure nickel, the quantity of iron present as an impurity being inmeasurably small. As the material is likely to become homogeneous by repeated annealing, the ovoids were carefully annealed for about 50 hours. The ovoid was wrapped in asbestus and placed in a thick metal tube, the interspace between the ovoid and the wall of the tube being filled with fine charcoal powder. The tube was then placed in charcoal fire. When the ovoid was annealed in this way, there were some traces of surface oxidation. The change of volume after each annealing was examined with the result that it became evident that the process of annealing increases the effect. It therefore appears that the previous history of the specimen exercises an important effect on the magnetization and on the dimensions of ferromagnetics as affected by magnetization. The anomaly in the length change noticed by Bidwell in two specimens of nickel wire is probably not the effect of temperature, but is perhaps to be ascribed to the cause above In contradiction to our former result with a square prism, the ovoid showed increase of volume. The amount of increase was small compared with the decrease noticed in the previous experiment. Cantone<sup>1)</sup> obtained a tolerably large increase of volume in nickel ovoids; our former result was nearly half as large, while in the present experiment, there is a slight increase. The discrepancy is probably due to the difference of treatment before the specimen can be converted into a proper shape for experimenting, and also to its chemical composition.

11. The volume change of ferromagnetics considered as a function of the magnetizing field takes place very slowly in weak fields; it then increases in a more rapid ratio till it reaches the

<sup>1)</sup> Cantone, Atti della R. Accad. die Lincei, 6, (1), 257, 1891.

'wendepunkt'; after that the change becomes slower, but still goes on increasing nearly in a straight line. Up to H=2000, the rate of change shows no tendency to decrease. With a still stronger field, the increase of volume will probably become more considerable.

12. In our former experiment, the range of the magnetizing force was confined to a few hundred C. G. S. units. In the present experiment, the increased field strength unveiled the character of the change of the volume considered as function of the intensity of magnetization. As will be seen from the curves (Fig. 2.) in dotted lines, the increase in nickel and steel takes place quite slowly before the magnetization reaches saturation. As soon as the magnetization reaches this state, the increase becomes very rapid, so that the branch of the curve ascends nearly parallel to the axis of volume increase. There we find that a slight increase in magnetization is attended with a large increase of volume. As the rate of increase appears to be nearly constant, it would be very interesting, if we could push the field strength still farther to see whether the volume change ultimately attains an asymptotic value.

The observed changes of volume and of length are exhibited in the following table:—

<sup>1)</sup> Cantone, Atti della R. Accad. dei Lincei, 6, (1), 257, 1891.

Nic	kel (1)	Soft	t iron (3)		8	teel (5)	Wolfr	am steel (8)
Н	$\frac{\partial v}{v}$	П	8r r	1	П	Öv v	Н	-ôv '
13	$0.09\times \bar{10}^7$	8	$0.10 \times 1$	-7 10	7	$0.08 \times 10^{7}$	19	$0.30 \times 10^{7}$
30	0.29	11	0.52		12	0.47	4:2	1.52
90	0.65	18	1.56		33	1.95	93	3.03
218	0.82	167	3.12		192	3.13	216	5.01
282	0.97	443	3.85	ì	376	4.69	44:2	8.04
517	1.38	691	4.58		586	6.22	692	11.68
877	2.06	958	5.88	i	792	8.01	1001	16.68
1141	2.44	1115	7.18	]	L()44	10.16	1117	18.96
1547	3.24	1342	9.47	]	1376	14.07	1296	22.75
1740	3.53	1563	11.45	1	1646	17.20	1704	28.82
2253	4.12	2089	14.68	1	2171	22.20	2153	32.62

Nickel (2)	Soft ir	on (4)	18	teel (6)	Wolfra	ım steel (8)
II 8/	H	81 1	Н	91	H	81 1
4 - 14.1×1	$\overline{\overset{7}{10}}$ 6	$2.5 \times 1$	[0] 13	$3.1 \times \overline{10}$	7 18	$4.1 \times \overline{10}^{7}$
6 - 64.0	1.5	19.0	19	7.1	2.5	12.4
10 - 118.2		31.6	28	15.1	39	21.7
15 = 163.6	127	23.8	.54	22.2	62	28.9
33 - 217.5	224	3.1	. 96	23.1	106	32.1
59 = 264.3	354 —	17.7	160	17.8	170	32.3
124 - 317.6	575 —	52.6	225	8.2	235	31.7
302 - 343.6	698 —	62.6	374	-11.5	349	30.2
561 = 353.8	883 —	73.5	589	-36.0	592	23.1
839 -356.0	1077 -	78.9	844	-49.2	781	18.7
1145 = 360.0	1180 -	82,2	1061	—.ĭ.ĭĭ	1052	17.0
1289 = 360.9	1324 -	86.6	1177	<b>-</b> 59.5	1188	15.4
1483 = 362.2	1447 —	89.9	1361	-64.5	1345	13.9
1849 - 362.7	1538 -	91.6	1729	=69.9	1697	12.4
2322 - 365.3	2180 =	102.0	2172	-78.1	2235	10.9

### Kirchhoff's Constants k' and k''.

13. Starting from the formulæ

$$\begin{split} \lambda &= \frac{\delta l}{l} = \left\{ \frac{4\pi k^2}{3} - \left( \frac{1+\theta}{1+2\theta} \right) + \frac{k-k'}{2(1+2\theta)} - \frac{k''}{2} \right\} \frac{H^2}{E}, \\ \text{and} \quad \sigma &= \frac{\delta v}{v} = \left\{ \pi k^2 + \frac{3(k-k')}{4} - \frac{k''}{4} \right\} \frac{H^2}{K(1+3\theta)}, \end{split}$$

which give the change of volume and of length of ferromagnetic ovoids in terms of Kirchhoff's constants k' and k'', we obtain the following expressions for these two constants:

$$k' = \frac{\rho(1+2\theta) - q}{2(1+3\theta)},$$
and 
$$k'' = \frac{3q - p}{2(1+3\theta)}.$$
where 
$$p = -\frac{4K(1+3\theta)}{H^2} \sigma + 4\pi k^2 + 3k,$$
and 
$$q = -\frac{2E(1+2\theta)}{H^2} \lambda + \frac{8\pi k^2}{3} (1+\theta) + k.$$

These constants, as calculated from the change of dimensions of ovoids, are given in the following table, and graphically drawn in Fig. 3:

Н	Nick	el	Soft	iron	St	eel	Wolfra	un steel
11	l.'	1:''	<i>l.</i> ·′	$k^{\prime\prime}$	<i>k'</i>	1:''	<i>l.</i> ·′	<i>l:''</i>
5	-229100	712800	21900	-22610	1017	-1865	348	-1252
10	-188900	578900	23520	-28450	3840	-3322	986	-1512
20	-71000	216700	13280	-16420	4248	-4615	3600	-3983
30	- 36370	111200	7302	<b>—</b> 8650	4048	-5080	4881	-5440
60	- 8163	34540	2139	- 2222	1738	-1864	1916	-2217
80	- 6906	20960	1207	-1102	1069	-1004	1198	-1385
100	-4653	14120	75)	- 550	701	-546	794	-880
120	- 3373	10260	500	-255	477	- 279	557	- 595
160	- 1297	3968	289	18	240	- 16	315	- 317
250	- 843	2553	5.5	175	69	117	128	-109
300	591	179)	25	175	:38	124	88	- 66
500	- 216	655	-9	130	— i	99	28	- 8
800	- 86	259	9	70	15	57	8	5
1200	- 89	117	-6	37	-4	31	::	.5
1600	- 22	65	<b>-</b> ‡	23	-3	19	1	4
2000	- 14	42	-3	16	-2	1:3	()	3

14. The curves for k' and k'' present the same general feature in iron and steel. "increases in flow fields; and there attaining the maximum value, it rapidly diminishes till it becomes less than zero; it then reaches a minimum, after which it again gradually increases. The exact position of the minimum is very vague; the curve for k' ultimately coincides with the axis of H. k'' is at first negative, and attaining the minimum value, goes on gradually increasing till it becomes greater than zero, and then reaches a maximum. With the farther increase of the field, the value of k" decreases very slowly. The position of maximum for k' and that of minimum for k'' lie nearly in the same field, which is greater for wolfram steel than for soft iron, while that for ordinary steel occupies an intermediate position. The absolute value of k' and k''is greater in iron than in steel. In nickel, the values of k' and k''are far greater than those for iron and steel, and moreover are of opposite signs. The maximum of k', or the minimum of k', seems to lie in a weak field; the rate of decrease or increase is quite rapid and the curves for k' and k'' soon approach the axis of H. Compared with the results of former experiments, the absolute values of k' and k'' are generally small for iron,—far greater for nickel. This difference arises from the fact that for iron, the change of length in weak fields is less in this case than in the former experiment, and that for nickel the contrary is the case. As regards the sign, these two experiments show fair agreement.

### Consequences of the theory.

15. Effect of longitudinal pull.—The change of magnetization produced by the clongation of a wire can be easily calculated from the formula

$$\partial \mathbf{I} = H \left\{ k' - \frac{E}{K} - 3 \left( k' + \frac{k''}{3} - \right) \right\} \lambda.$$

Putting  $\lambda = 4.67 \times 10^{-6}$ ,  $4.80 \times 10^{-6}$ , and  $4.85 \times 10^{-6}$  for soft iron, ordinary steel, and wolfram steel respectively, each corresponding to a pull of 0.1 Kilog. per sq. mm., we get the following results:

II	Soft iron.	Steel.	Wolfram steel.
11	ôΙ	$\partial I$	$\partial I$
10	0.919	0.055	(1.044
20	1.074	0.212	0.170
30	0.831	0.402	0.350
60	0.399	0.2.54	0.294
80	0.244	0.153	0.249
100	0.127	0.072	0.188
120	0.039	0.005	0.14.5
160	-0.080	-0.092	0.094
200	-0.164	-0.146	0.061
300	-0.25S	-0.210	0.017
500	-0.311	-0.236	-0.002
800	-0.275	-0, <u>2</u> 0.5	-0.038
1200	-0.219	-0.163	-0.037
1600	-0.183	-0.134	-0.033
200)	-0.157	-0.115	-0.028

It will be seen from the above table that there is an increase of magnetization in low fields, till it reaches a maximum, after which it gradually decreases. The decrease does not proceed continuously, but reaches a maximum, whence the magnetization begins to recover. Although the former result here arrived at is the well-known Villari effect, we do not know whether the maximum decrease due to longitudinal stress has as yet been experimentally ascertained. With nickel, we obtain the following values for the change of magnetization due to clongation,  $\lambda$ =

$4.74 \times 10^{-6}$ , which	corresponds to	a pull of 0.1	Kilog. per sq. mm.:
-------------------------------	----------------	---------------	---------------------

11	$\partial I$
10	-24.58
20	-18.87
30	-14.16
60	- 9.09
80	- 7.12
100	- 5.99
120	- 5.22
160	- 2.70
300	- 2.28
500	- 1.39
800	- 0.88
1200	= 0.60
1600	- 0.45
2000	- 0.36

There is nothing remarkable in nickel. Longitudinal pull produces decrease of magnetization, which becomes gradually less as the field strength is increased. This is such a well established experimental fact that we need not enter into further discussion of the subject.

16. Effect of hydrostatic pressure.—We can easily see that the change of magnetization  $\partial I$  due to change of volume  $\sigma$  by hydrostatic pressure is given by

$$\partial \mathbf{I} = - \bigg( \, k' + \frac{k''}{3} \, \bigg) H \sigma \; . \label{eq:deltaI}$$

If we calculate the change of magnetization due to contractions  $4.68 \times 10^{-6}$ ,  $5.38 \times 10^{-6}$ ,  $8.42 \times 10^{-6}$  and  $9.33 \times 10^{-6}$  for nickel, soft iron, steel, and wolfram steel respectively, each corresponding to a pressure of 10 atm., we obtain the following values:

Н	Nickel	Soft iron	Steel	Wolfram steel
11	$\partial I$	$\partial I$	$\partial I$	òΙ
0	0.000	0.000	0.000	0.000
10	0.190	0.757	0.230	0.045
20	0.119	0.840	0.457	0.237
30	0.080	0.713	0.595	0.858
60	0.037	0.398	0.565	0.675
80	0.030	0.362	0.495	0.550
100	0.024	0.305	0.437	0.467
200	0.012	0.178	0.268	0.259
300	6.008	0.135	0.200	0.185
500	0.005	0.093	0.134	0.118
800	0.002	0.062	0.088	0.073
1200	100.0	0.042	0.061	0.048
1600	0.000	0.031	0.043	0.034
2000	0.000	0.024	0.034	0.026

It thus appears that in nickel the effect of hydrostatic pressure is very small compared to that of longitudinal pull. There is increase of magnetization with the volume contraction of the magnet. Such an increase reaches a maximum in low fields, whence the effect gradually diminishes. Similar changes are also noticed in the case of iron and steel. In our former experiment, we found that hydrostatic pressure increases the magnetization in nickel, while it decreases it in iron. The agreement between theory and experiment is very close in nickel, but there is a wide discrepancy in iron and steel, as we have already noticed.

17. Effect of torsion on longitudinally or circularly magnetized wire. There are other important consequences to be drawn from the constant k" with regard to the effect of torsion on

longitudinally magnetized wire and on ferromagnetic wire traversed by an electric current. The strain caused by twisting a circular wire can be resolved in elongation and contraction in directions perpendicular to each other and inclined to the axis of the wire at 45°. Taking these two principal axes of the strain for those of x and y, we have for the strain.

$$\frac{\partial u}{\partial x} = \frac{1}{2}\omega r,$$

$$\frac{\partial v}{\partial y} = -\frac{1}{2}\omega r,$$

$$\frac{\partial w}{\partial z} = 0,$$

where  $\omega$  denotes the amount of torsion and r the distance from the axis. Resolving the magnetizing force which is in the direction of the axis of the cylinder, along the axis of elongation and of contraction, we find that the circular magnetization which will be called into play is equal to  $-\frac{1}{2}\omega rk''H$  at a distance r from the axis, the mean circular magnetization being  $-\omega k''HR$ , where R is the radius of the wire.

The transient current which will be thus induced in the wire by suddenly twisting it is proportional to -k''H.

Next suppose that the wire is traversed by an electric current of intensity C. Then the circular magnetizing force at a distance r from the axis is

$$H = \frac{2C'r}{R^2}$$
.

By applying similar reasoning, we find that the mean longitudinal magnetization is equal to  $-\omega k'' C'$ . We therefore conclude that twisting the wire earrying the electric current gives rise to longitudinal magnetization proportional to -k'' C. Thus the circular magnetization produced by twisting a longitudinally

magnetized wire has a reciprocal relation to the longitudinal magnetization caused by twisting a circularly magnetized wire.

The view propounded by Prof. Ewing<sup>2)</sup> to account for the existence of transient current by means of avolotropic susceptibility is similar to what would follow from Kirchhoff's theory, but it fails to give the amount of the current or of the magnetization which would be produced by twisting.

The theoretical inferences which we can draw at a glance from the curves of -k''H (Fig. 3) are as follows:

- 1. The transient current as well as the longitudinal magnetization produced by twisting an iron or steel wire is opposite to that produced by twisting one of nickel, up to moderate fields.
- 2. The transient current as well as the longitudinal magnetization produced by twisting an iron, steel, or nickel wire reaches a maximum in low fields.
- 3. In strong fields the direction of the current as well as the longitudinal magnetization is the same in iron, steel, and nickel.

It has been established by G. Wiedemann<sup>3</sup> that the longitudinal magnetization produced by twisting an iron wire carrying an electric current is opposite to that produced in a nickel one. The opposite character of the transient current in these two metals has also been observed by Zehnder<sup>1</sup> and independently by one of us<sup>3</sup>. The existence of a maximum transient current in

<sup>1)</sup> Voigt, Kompendium der theoretischen Physik, 2, 200, 1896, Leipzig: Drude, Wied. Ann. 63, 8, 1897.

<sup>2)</sup> Ewing, Proc. Roy. Soc. 36, 1884.

<sup>3)</sup> Wiedemann, Electricität, 3.

<sup>4)</sup> Zehnder, Wied. Ann., 38, 68, 1889.

Nagaoka, Phil. Mag. [5] 29, 123,1890; Journal of the College of Science, Tōkyō, 3, 335, 1890.

these two metals has been clearly established, although there is some difference in the field strength between iron and nickel. It appears from the experiments of Dr. Knott<sup>1)</sup> that the area of the hysteresis curve in the longitudinal magnetization produced by twisting circularly magnetized wire reaches a maximum as the field strength is increased; but on account of the feebleness of the current, the existence of the maximum in the longitudinal magnetization is not well established. To judge from the course of the curve given by the same experimenter, it seems highly probable that the maximum would be reached if we could push the circularly magnetizing force a little further. The conclusion (3) is still an open question, although some experiments of Matteucci<sup>2)</sup> seem to corroborate the view just stated.<sup>3)</sup>

18. Looking at the curves of k"II, we cannot but be struck with the close resemblance of the curves representing the amount of torsion produced by the combined action of the circular and the longitudinal magnetizing forces on a ferromagnetic wire. We can no doubt co-ordinate the effect of torsion on a magnetized wire with the Wiedemann effect. The discussion of the last mentioned effect we hope to lay before the public in the near future.

In spite of the qualitative explanations which Kirchhoff's theory affords with regard to the effect of longitudinal pull, of the hydrostatic pressure, and of torsion, there are instances in which the theory apparently fails in several quantitative details that it necessarily calls for modification. We may remark that k'

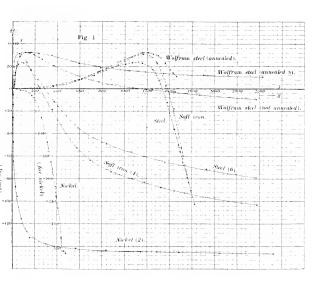
<sup>1)</sup> Knott, Trans. Roy. Soc. Edinb., 36, 485, 1891.

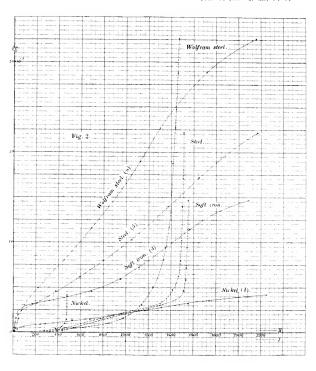
<sup>2.</sup> Matteucci, Annales de Chimie et de Physique, 1858.

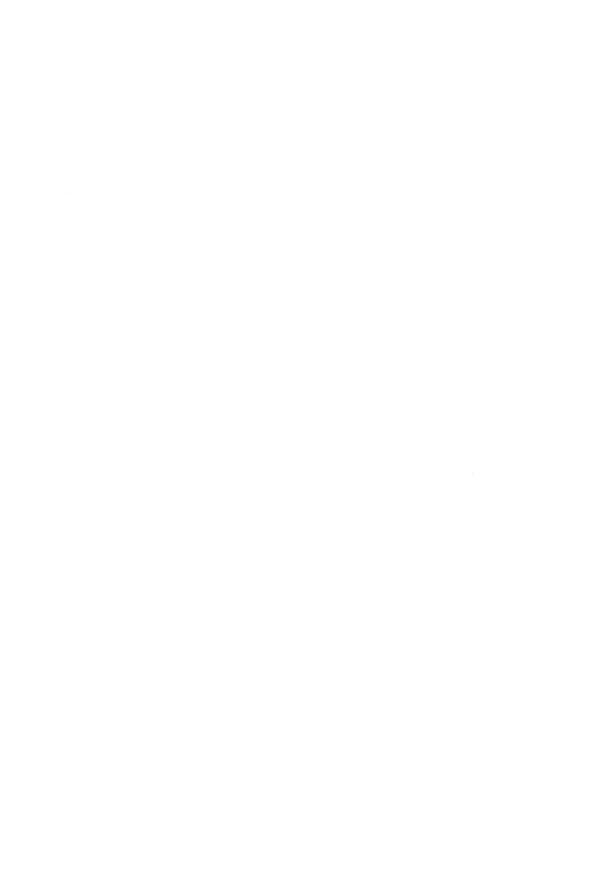
<sup>3)</sup> While this paper was passing through the press, we found that the direction of the transient current produced by twisting a magnetized iron wire is reversed in strong magnetizing fields.

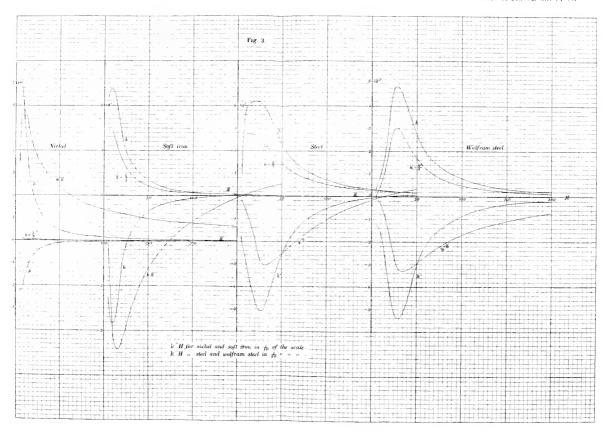
and k" are physically functions of the strain, as is borne out by the numerous experiments on the effect of stress on magnetization. The present state of the theory of magnetostriction may perhaps be compared with that stage in the history of the theory of magnetization when the intensity of magnetization was supposed to be simply proportional to the magnetizing force. In fact, the theory is still in its infancy, so that there are ample grounds for expecting further developments on further researches.











# Combined Effect of Longitudinal and Circular Magnetizations on the Dimensions of Iron, Steel and Nickel Tubes.

By

**K. Honda**, *Rigakushi*, Post-graduate in Physics.

With Plates VIII. and IX.

1. The change of length in the direction of magnetization has been made a subject of investigation by several experimentalists, but few of them have measured the change in the direction perpendicular to that of magnetization. Joule<sup>1)</sup> first observed the diminution of length of an iron gas-piping by passing a current through an insulated wire inserted into it, and bent over the sides, so as to form a circular magnetizing coil of 1½ convolutions. His experiment was modified by Bidwell<sup>2)</sup> who measured the change of dimensions in an iron ring. He found that the ring becomes thicker in a strong field and thinner in a weak one. From the measurement of the internal as well as the external change of volume for iron, steel, nickel, and cobalt tubes, Knott<sup>3)</sup>

<sup>1)</sup> Joule, Scientific papers I, 263.

<sup>2)</sup> Bidwell, Proc. Roy. Soc. 56, 94, 1895.

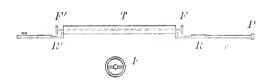
<sup>3)</sup> Knott, Trans. Rov. Soc. 39, 457, 1898.

calculated the change of lateral dimension in these tubes. The result for iron coincided qualitatively with that of Bidwell.

The first experiment on the change of length of an iron wire by the combined action of longitudinal and circular magnetizations was made by Beatson<sup>1)</sup> who observed the diminution of length at the moment when an electric current was passed through a magnetized wire. A similar result was afterward obtained by Righi.<sup>2)</sup> The same experiment was also repeated by Bidwell,<sup>3)</sup> who observed a large increase in the change of length by longitudinal magnetization of an iron wire carrying a current.

2. Through the kindness of Prof. Nagaoka, his apparatus<sup>4)</sup> for the [measurement of the minute change of length was placed at my disposal. The apparatus consists of a small optical lever with an arrangement for temperature compensation on the same principle as the gridiron pendulum. The rod, by which the change of length is made sensible to the lever, was slightly modified.

In the annexed figure, T is the tube to be tested, F and F'



are two circular brass rings Pprotruding from the tube
at a distance of 1 cm. from
the ends, and soldered to a
brass rod passing through

the axis of the tube. The magnetizing coil was wound round the tube parallel to its length extending from F to F' to envelope it completely, and so arranged that the tube could slide in the coil with little friction. F in the lower part of the figure shows

<sup>1)</sup> Beatson, Archives des. Sc. phys. et nat. 2. 113, 1846.

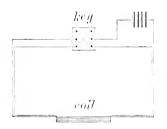
<sup>2)</sup> Righi, Mem. di Bologna 4, 1, 1879; Beibl. 4, 802.

<sup>3)</sup> Bidwell, Proc. Roy. Soc. 51, 495, 1892; Beibl. 17, 582.

<sup>4)</sup> Nagaoka, Phil. Mag. 27, 131, 1894; Wied. Ann. 53, 487, 1894.

the front view of these rings. R and R' were two rods in contact with the ends of the tube. The ends of these rods were bent upwards and so filed down, that they could easily slide between two parallel wires of the coil, which were specially fixed at a distance of 1 mm. from each other. The rod r served to communicate the motion to the prism P. The other parts of the apparatus remained unchanged. The apparatus was put into a magnetizing coil, 30 cm. long and wound in 12 layers with copper wire of 2 mm. diameter. The field at the centre of the coil due to a current of one ampere was 37.97 C.G.S. units. The current through the outer coil produced the change of length by longitudinal magnetization and that through the inner coil gave rise to the change of length by circular magnetization.

To study the effect of temperature on the change of length, the circular magnetizing coil was wound, not by a single wire, but by double wires; thus connecting the four ends of these



wires to a reversing key as shown in the figure, the circular field can be made or annulled by turning the key one way or the other. The total number of turns of the circular magnetizing coil was 44 for the nickel

tube, 40 for the wolfram steel tube and 36 for the soft iron tube. The magnetizing currents were measured by Thomson graded galvanometers which were compared with a deciampere balance before each experiment.

3. The samples used in the present experiment had the following dimensions:

material	length. (cm.)	external diam. (cm.)	internal diam. (cm.)	demagnetizing factor.
niekel	17.02	1.328	1.252	0.0261
wolfram steel	20.30	1.124	1.048	0.0162
soft iron	16.97	0.966	0.842	0.0308

The tubes of nickel and soft iron are the same as that used in the study of the mutual influence between longitudinal and circular magnetizations. It was found by analysis that the nickel was nearly chemically pure, the trace of impurity being inmeasurably small.

# Results of Experiments.

## 1. Nickel Tube.

4. The tube was carefully annealed, before the circular magnetizing coil was wound round it. The change of length due to longitudinal field alone was then measured in the usual way. The results were compared with that obtained after the circular magnetizing coil was wound round the tube. The comparison showed that there was in general small difference between these two, and that the change of length in the former case was always greater than that in the latter, the difference amounting to nearly 2 or 3 %. This is evidently due to the resistance to contraction experienced by the tube, although it can easily slide along the coil. Whether the apparatus executed its function correctly or not was tested before each experiment by making a longitudinal field and comparing the deflection so obtained with that in the free state; otherwise serious mistakes would sometimes have arisen.

5. The experiments on the change of length by circular magnetization, namely, on the change of dimension in a direction perpendicular to the magnetic field, were conducted in the following manner. The tube was first demagnetized and a circular magnetizing current was made only for a moment and the corresponding deflection read. The change of length due to magnetization followed almost instantaneously, but the change due to the heating of the coil became sensible somewhat later; hence these two effects were unmistakably distinguishable so long as the magnetizing current was not strong. On this account the highest field did not exceed 100 C.G.S. units.

The effect of the longitudinal field on the change of length by circular magnetization was also measured. A constant longitudinal field was first made and the corresponding deflection observed; then currents of different strength were momentarily passed through the circular magnetizing coil, and the additional deflection was read. These results are given in the following table and also in Fig. 1:

TABLE I.

Н	H=0		=6.9	H = 22.1	H=	:182.9
h	$\frac{\partial l}{l} \times 10^{7}$	h	$\frac{\delta l}{l} \times 10^{7}$	$h = \frac{\delta t}{t} \times 10^7$	h	$\frac{\delta l}{l} \times 10^{5}$
7.6	4.4	8.2	2.7	8.2 0.5	8.5	1.6
16.1	19.0	17.5	40.8	17.5 18.0	13.6	2.2
26.3	46.2	31.1	87.1	31.2 - 76.2	21.3	4.4
40.6	75.1	40.9	110.4	40.9 124.9	31.5	9.2
50.5	92.5	49.9	130.6	50.2 157.8	49.0	27.2
64.5	108.8	57.5	141.4	64.2 - 190.4	64.2	59.9
72.1	117.0	72.1	163.2	70.9 201.3	69.5	81.6
\$3.2	119.7	81.7	168.6	78.8 217.7	87.6	130.6

Here H and h denote effective longitudinal and circular fields respectively, both in C.G.S. units.  $\frac{\partial l}{l}$  represents additional change of length by circular field. All these changes were measured at a constant temperature of about 25° C.

Fig. 1 shows that the change of length by circular magnetization increases at first showly and then rapidly. With the further increase of the circular field, the rate of increase becomes gradually less. This result agrees in quality with Knott's calculation. The circular magnetization combined with a constant longitudinal one is always to increase the length which is first shortened by the longitudinal magnetization. In weak circular fields, the curve of the change of length with a constant longitudinal field lies below the curve with no such field; but in strong fields, the first curve lies above the second. The point of intersection of these two curves is displaced into a higher field with the increase of the longitudinal.

6. We shall next pass on to the change of length by longitudinal magnetization with a constant circular field. The tube was first demagnetized by reversals, and then the deflections for longitudinal magnetizing currents of different strength were measured. During the experiment, the temperature at the centre of the magnetizing coil was 18.8° C. The tube was then carefully demagnetized both as regards the longitudinal and circular magnetizations. Then a constant current was passed through the circularly magnetizing coil so that the field strength became null. Owing to the heating of the coil, the tube rapidly expanded at first, but usually after an hour or two, it reached a stationary state; when that state was reached, the measurement of the change of length by longitudinal field alone was commenced, which gave the length change at a higher temperature. After

the observation was finished, the key was reversed, so that the circular magnetizing current was then called into play. During this process, no gradual displacement was observed, showing that the temperature of the tube remained unchanged during the reversal, but at the same time an instantaneous deflection was noticed, which showed the change of length by circular magnetization. By reading the displaced position of the line in the micrometer ocular, the deflection corresponding to the longitudinal magnetization was noted. The tube was then demagnetized as regards the longitudinal magnetization, the circular magnetization remaining constant. The same process was repeated for stronger fields, till a set of observations was completed.

7. How the rise of temperature affects the change of length by magnetization will be seen from Fig. 2. The change of length at ordinary temperature is somewhat less than that which Prof. Nagaoka and myself 1) have obtained for an ovoid made of the same specimen. The difference may perhaps be explained by that of annealing and of the geometrical shape of these samples. The temperature was measured by inserting a mercury thermometer inside the tube. Its effect is thus tolerably large; the rise of temperature is attended with an increase of the change of length in weak fields, and is accompanied with a decrease in strong fields. From the same figure, we obtain the relation of temperature to the change of length at a constant field as shown in Fig. 3. It is well known that the magnetization of nickel increases with temperature in low fields and decreases in strong ones; but under the temperature of 100° C, the change of magnetization is too small to account for the change of length.

<sup>1)</sup> Nagaoka and Honda, Preceding paper.

So far as I am aware, Barrett<sup>1)</sup> is the only physicist who has investigated the effect of temperature on the change of length; his experiment resulted in the decrease of about one-fourth of the change of length by a rise of temperature by about 50° C. Perhaps his field was too strong to cause an increase.

S. The results of the change of length by longitudinal magnetization with a constant circular field are given in the following table and in Fig. 4. The change of length was reduced to the temperature of 18.8° C by using the results above obtained.

TABLE II.

h	=0	h=	=10.7	10.7 h=16.8		h:	=22.9
Н	$\frac{\delta l}{l} \times 10^7$	Н	$\frac{\delta l}{l} \times 10^7$	11	$\frac{\partial l}{l} \times 10^7$	Н	$\frac{\delta l}{l} \times 10^7$
5.3	- 1.5	6.9	- 10.9	8.1	- 17.6	7.2	- 13.0
8.5	- 25.6	14.6	-61.7	17.8	<b>-</b> 73.5	14.2	- 49.4
17.0	-71.7	27.8	-118.1	27.4	-118.1	22.3	<b>-</b> 93.9
29.4	-107.6	42.7	-161.6	46.7	-174.9	37.6	-149.2
41.2	<b>-</b> 134.7	62.8	-198.6	71.9	-223.4	65.1	-198.7
61.0	<b>-163.9</b>	103.3	-238.4			94.0	-232.7
101.3	-202.4	131.6	-255.2	131.9	-271.0	129.7	-269.3
184.9	-241.8	176.8	-279.2	177.7	-291.5	175.2	-293.4
274.9	-261.3	254.0	-301.7	255.4	-313.7	255.5	-317.0
354.9	-274.1	361.0	-312.9	363 4	-329.7	359.3	-333.2
468.6	-279.2	505.0	- 324.0	516.1	-339.4	516.1	-347.4
709.2	-289.5	720.2	-331.6	779.0	-344.5	725.2	-351.4

<sup>1)</sup> Barrett, Phil. Mag. [4] 47, 51, 1874; Nature 26, 515-586, 1882; Beibl. 7, 201.

The comparison of Figs. 1 and 4 shows that the general character of the change of length is the same in these two cases, except that the sign of the change is opposite. Hence similar remarks as in the former hold good in the present case.

In the experiment with nickel and cobalt wires traversed by an electric current, Bidwell found that the effect was inmeasurably small. The discrepancy in nickel perhaps arises from the effect of temperature, which he did not take into account; the difference in the method adopted in the present experiment for obtaining a circular field and in that of Bidwell does not seem to play an important part in accounting for the said discrepancy. According to the present experiment, the rise of temperature occasioned a comparatively large diminution of the length change in strong fields. Hence it can not be denied that in Bidwell's experiment, the effect of circular magnetization was just as great as that of temperature. The same remark will perhaps apply to his experiment with cobalt; but having no cobalt tube at my disposal, the experimental verification must be postponed till some However, a theoretical deduction in favor of the future date. view above stated will be given in the last part of the paper.

It would not be out of place to remark that a klinging note of the nickel tube was heard at the make and break of circular magnetizing current, a well known phenomenon. Even with such a weak current as we obtain from a single Daniell's cell was sufficient to produce a distinctly audible sound.

9. It will sometimes happen that it is convenient to have a simple expression for the change of length. For nickel, the change of length is very well given by an empirical formula of the form

$$\frac{\partial l}{l} = -\frac{\alpha H^n}{1 + \beta H^n},$$

where  $\alpha$ ,  $\beta$  and n are constants and H is assumed to be positive. The determination of these constants from the experimental curve gave the following results:

$$\alpha = 5.18$$
,  $\beta = 0.0164$  and  $n = 1.017$ .

In the calculation, only the fields H=20, 80, 320 were chosen to simplify the calculation. Using these values of the constants, the change of length due to fields of different strength was calculated and compared with the experimental value as shown in the following table:

TABLE III.

Н	$-\frac{\partial l}{l}$ (cal.)	$-\frac{\delta l}{l}$ (exp.)
10	$-46 \times 10^{-7}$	- 40×10 <sup>-7</sup>
20	- 81	- 81
30	-108	-109
50	148	-148
80	-185	-185
120	-215	-214
1.50	-230	-229
200	-247	-246
250	-258	-258
320	-269	-269
400	-278	-278
500	-285	-284
600	-292	-289
700	-293	-291

Thus except in weak fields, the coincidence between these two is very close; the difference does not amount to 1%. This formula applies, not only for the change of length, but also for every curve which has only one inflexion point and becomes asymptotic when one of the co-ordinates increases indefinitely, such as the curve of magnetization.

#### 2. Wolfram Steel Tube.

10. The method of procedure with the steel tube was exactly the same as in the corresponding case of the nickel tube. The result of the change of length by circular magnetization, e.g., the dilatation in a direction perpendicular to the field, as well as the effect of longitudinal field on the change of length by circular magnetization are given in the following table and graphically shown in Fig. 5. These observations were taken at a constant temperature of about 17° C.

TABLE IV.

H = 0	H=0 $H=15.1$		II = 81.3	
$h = \frac{\delta l}{l} \times 10^7$	$\frac{h}{l} \times 10^7$	$h = \frac{\epsilon_l}{l} \times 10^7$	$\frac{1}{l} \times 10^7$	
14.0 - 0.0	13.2 - 0.4	13.6 - 0.4	12.9 - 0.4	
20.8 - 8.6	31.7 - 12.9	30.9 - 2.1	31.2 - 0.9	
35.2 = 20.2	41.9 - 32.2	41.9 - 14.6	41.6 - 5.2	
51.8 - 22.8	51.7 - 40.4	51.1 - 25.8	51.1 - 12.9	
65.1 - 26.6	63.7 - 45.1	63.7 - 38.7	63.7 - 25.8	
78.7 - 27.9	74.7 - 47.3	74.7 - 48.1	75.6 -30.9	
99.5 = 27.9	88.6 -49.4	91.4 - 58.0	88.6 -40.0	

88 K. HONDA.

Here we observe that circular magnetization produces contraction which increases very slowly at first, but afterwards quite rapidly, till it reaches a nearly constant value. The existence of the field of maximum contraction is still a question. The result is somewhat discordant as compared with that of Bidwell with an iron ring, in which case the diminution vanishes in a field of about 86 C. G. S. units. Since the behaviour of wolfram steel as regards the change of dimensions by magnetization is very different from that of soft iron, the cause of the discrepancy is probably to be sought for in the difference of the specimens.

That the effect of longitudinal field on the change of length by circular magnetization is of the same nature as in the case of nickel, except that the sign of the change is opposite, is also apparent from the same figure. As we have remarked, Beatson and Righi observed the same phenomenon.

11. The middle curve in Fig. 6 represents the change of length by longitudinal magnetization at the temperature of the room. The lower curve was obtained at 80.2° C, and the upper curve at the same temperature by reversing the key so as to produce circular field. From the figure, we see that the behaviour of wolfram steel as regards the change of length is widely different from that of other sorts of iron. It is remarkable that the length of the tube, after reaching the maximum elongation, diminishes very slowly as the field is increased, a fact already noticed by the experiment referred to. In that case, the maximum elongation was somewhat less than in the present experiment. The discordance between the two is probably due to the difference of annealing and also of the shape of the specimens.

<sup>1)</sup> Nagaoka and Honda, loc. cit.

The effect of temperature is to decrease the change of length; the diminution increases with the field, till it reaches a maximum, and then decreases very slowly. Barrett<sup>1)</sup> did not find the effect in the case of iron and cobalt. The upper curve shows that the influence of circular magnetization on the change of length is large for steel.

12. The effect of circular field on the change of length by longitudinal magnetization is shown in the following table and in Fig. 7. The results are reduced to the temperature of 17.2° C.

h	h = 0 $h = 10.8$		0.8	h=	17.7	h = 25.8	
II	$\frac{\tilde{c}l}{l} \times 10^7$	Н	$\frac{3l}{l} \times 10^7$	Н -	$\frac{\delta l}{l} \times 10^7$	Н	$\frac{\delta l}{l} \times 10^7$
12.0	0.0	19.3	3.7	14.5	0.2	11.3	0.0
17.7	8.3	30.5	18.3	26.1	11.9	21.0	2.1
29.3	16.2	37.6	24.5	31.9	18.0	31.2	17.2
49.4	27.3	53.1	35.8	50.8	37.2	57.7	41.8
93.0	38.8	84.6	46.3	75.7	50.5	83.7	56.8
125.1	42.9	135.3	52.4	105.5	54.5	120.3	67.6
170.0	44.9	182.5	54.2	168.4	60.1	165.8	72.5
348.5	43.3	233.3	55.0	244.5	63.7	246.5	75.2
441.0	42.9	325.5	55.6	350.0	63.9	351.5	73.3
545.0	42.5	505.0	54.2	461.5	63.6	459.5	72.3
728.0	41.3	708.8	52.4	615.5	62.8	671.6	71.0

TABLE V.

Thus the longitudinal magnetization combined with a constant circular one is always to increase the length which is first

<sup>1)</sup> loc. cit.

shortened by the circular magnetization. In weak longitudinal fields, the curve of the change of length with a constant circular field lies slightly below the curve with no circular field; but in strong fields, the first curve lies markedly above the second. The point of intersection of these two curves shifts into a high field as the circular field is increased. The field of the maximum elongation seems to increase with the circular field.

## 3. Soft Iron Tube.

13. The experiments of the change of length by circular magnetization and of the effect of longitudinal field on the change of length led to the following results, which are graphically shown in Fig. 8. The observations were taken at the temperature of 18° C.

TABLE VI.

H=0	H = 5.7	H = 25.8	H = 67.6
$\frac{1}{\ln -\frac{\delta l}{l} \times 10^7}$	$h \frac{\partial l}{\partial r} \times 10^7$	$h = \frac{\delta l}{l} \times 10^7$	$h = \frac{\delta t}{t} \times 10^7$
5.3 - 7.8	5.3 - 4.2	5.3 - 0.0	5.3 - 0.5
14.0 -13.0	13.8 - 11.9	14.0 - 5.2	13.8 - 1.0
21.4 - 15.6	20.7 - 16.6	21.4 - 10.4	21.0 - 4.2
37.5 -15.6	35.7 - 20.8	37.3 -20.8	37.3 - 9.9
53.2 - 14.5	51.8 -22.3	53.3 -26.0	53.2 -14.0
69.4 - 12.5	66.6 -22.3	69.2 -28.0	67.8 $-18.2$
81.6 - 9.3	81.3 -20.8	81.6 - 26.0	80.5 -20.8
98.8 - 7.8	97.7 - 19.7	98.0 -23.4	98.1 -20.8

By circular magnetization, the length of the tube diminishes rapidly at first, till it reaches a minimum, then it gradually recovers. The field at which the tube returns to its former length is not yet reached so far as the present experiment extends. The result agrees qualitatively with that of Bidwell and the calculation of Knott.

The general form of the curve does not change by the application of a constant longitudinal field, but the field of maximum contraction shifts into high field as the longitudinal field increases. The amount of the maximum contraction increases with the longitudinal field, till it reaches a maximum, and then it gradually decreases. In weak circular fields, the change of length diminishes with the increase of the longitudinal.

14. As in the case of wolfram steel, three curves in dotted lines are given in Fig. 9, two of which correspond to the change of length at the temperatures of 18.7°C and 76.1° respectively. When the key in the circuit of the circularly magnetizing coil was reversed so as to produce a field, the change of length corresponding to the third curve was obtained.

The change of length by longitudinal magnetization at ordinary temperature is somewhat less than those obtained by previous experimenters. The difference is probably to be ascribed to the well annealed state<sup>11</sup> of the tube; also, the resistance to the elongation experienced by the tube due to the friction of the circular magnetizing coil was found to affect the result slightly. The general feature of the change of length is so well known that farther remarks are unnecessary. It is only to be noticed that here the field of the maximum elongation is greater by 20 C. G. S. units than that of the minimum contraction due to circular magnetization.

The rise of temperature is to diminish the change of length

<sup>1)</sup> Bidwell, Phil. Mag. 55, 228, 1894.

92 K. HONDA.

in weak fields and to increase it in strong ones. The field at which the temperature produces no effect is about 52 C. G. S. units. In the case of wolfram steel, this field, if it exists, seems to be pushed to an intensely strong field. We also observe that the effect of circular field on the change of length by longitudinal magnetization is tolerably large, as observed by Bidwell.

15. The results of the experiments on the change of length by longitudinal magnetization with a constant circular field are summed up in the following table and graphically shown in Fig. 10, these results being reduced to 18.7° C.

h = 5.7h = 26.2h = 0h = 9.2 $\frac{\delta l}{r} \times 10^7$  $^{50}$   $\times 10^{7}$  $^{5l}_{-1} \times 10^{7}$  $\frac{\delta l}{l} \times 10^7$ Н H Н Η 5.3 1.1 6.9 4.7 5.3 1.1 5.3 1.7 10.3 12.8 11.217.210.3 9.6 17.117.9 22 7 21.5 22.4 22.9 29.6 20.541.3 19.2 37.8 28.8 39.4 37.4 40.6 35.6 70.3 19.261.240.0 61.8 32.164.0 42.397.9 17.1111.1 31.0 97.9 42.2 90.9 41.1 144.3 11.1 38.8 38.2142.627.8 145.0143.5223.0 3.6 217.234.5218.021.4217.631.0 318.5-4.912.4 320.0 20.2312.525.3320.3 481.4 -20.34.2 475.0 8.6 490.0 -4.3493.8 -2.6697.0 -32.5704.0 -13.5684.2 -5.7647.0

TABLE VII.

Thus the nature of the change of length is the same as in the reciprocal case already mentioned, except that the sign of the change is opposite. As shown in the figure, in strong fields, the curve corresponding to the change of length with a constant circular field lies always above that with no circular field. In weak fields, the curves nearly coincide with each other. The field of maximum elongation slightly increases with the circular, and the amount of the elongation, after reaching a maximum, begins to decrease with further increase of the circular field. Though Bidwell did not observe this point, the present experiment agrees quite well with his result.

16. So far the experiments made on the tubes of nickel, steel and iron show that the effect of circular field on the change of length by longitudinal magnetization is of the same nature as the effect of longitudinal field on the change of length by circular magnetization.

From the results of the change of length by longitudinal and circular magnetizations, the change of volume by magnetization can easily be calculated, provided we assume the material to be isotropic, as was already done by Bidwell. If u and v represent these two dilatations respectively, the volume change  $\sigma$  is given by the formula  $\sigma = u + 2v$ .

Assuming the isotropy of our specimens, we find the calculation leads to the following results:

	Nickel	Wolfram steel	Soft iron
IŦ	$\frac{\partial v}{v}$	-	- <u>&amp;v</u>
10	$-18.5 \times 10^{-5}$	$0.0 \times 10^{\circ}$	$-\frac{1}{2}9.6 \times 10^{7}$
20	<b>-21.</b> 0	- 7.2	-13.1
30	0.0	-18.8	-12.8
40	18.0	-22.2	-12.2
60	46.5	-21.8	- 7.7
80	.54.0	-19.2	<b>-</b> 1.9
100		-16.4	1.5

TABLE VIII.

94 K. HONDA.

We thus obtain incredibly large values for the change of volume. In nickel and soft iron, there is at first decrease of volume, and then follows an increase; in wolfram steel, the diminution of volume reaches a maximum and then gradually decreases. The above result for soft iron agrees fairly well with that of Bidwell<sup>1)</sup> for unannealed iron ring. But in the experiment with ovoids<sup>2)</sup> made of the same specimens, there was always small increase of volume for nickel, steel and soft iron. The amount of the change at the field of 100 C. G. S. units was  $0.7 \times 10^{-7}$ ,  $3.1 \times 10^{-7}$ and  $2.8 \times 10^{-7}$  for these metals respectively. Hence the question now arises whether the change of volume is so influenced by the shape of these metals. To settle this point, fresh experiments on the change of volume were undertaken with a dilatometer. answer was in the negative, the result being in rough agreement with that for the ovoids. The initial decrease of volume was never observed, but the volume always increased with the increase of the magnetizing field. The discrepancy between the calculated and the experimental result is perhaps due to the evolutropy of the materials. For, if it were not isotropic, the lateral dilatation by longitudinal magnetization would not coincide with the change of length by circular magnetization. It will also be explained by the ecolotropy of the specimens that in weak fields, Bidwell's calculation resulted in the large diminution of volume of iron rings in contradiction to the experimentally established fact.

loe, cit.

<sup>2)</sup> Nagaoka and Honda, loc. cit.

# Concluding Remarks.

17. From the experiment on the relation between magnetism and twist, Knott<sup>1</sup> concluded that the pure strain effects on a ferromagnetic wire caused by tension and longitudinal current through it are of an opposite character, and also, on the ground of Maxwell's explanation for Wiedemann's effect, that in an iron or nickel wire carrying an electric current, the change of length by magnetization must be greater than when there is no longitudinal current. Since the change of length for cobalt is scarcely affected by tension, the same must also be the case for longitudinal current. The consideration is partially verified by the experiment of Bidwell and also by the present one.

The same phenomenon may also be more concisely explained in the following manner. Suppose our samples to be isotropic and to have no residual effect. Let l and t be two magnetic forces acting longitudinally and circularly in two perpendicular directions. When these two forces act simultaneously, we have a resultant force H: this force occasions the change of dimensions in our ferromagnetics. The dilatation in the direction of the resultant force, as well as that in the direction perpendicular to it, can be expressed by f(H) and F(H) respectively, which are even functions of H. To obtain the dilatation in the longitudinal direction, we have simply to construct a strain ellipsoid at any point of the ferromagnetics and to find the change of length of the radius vector in this direction. The simple calculation gives

$$\frac{\partial L}{L} = f(H) \frac{\ell^2}{H^2} + F(H) \frac{\ell^2}{H^2}.$$

<sup>1)</sup> Knott, Trans. Roy. Soc. Edinb. 36, pt. II., 485.

In the case of nickel, the change of volume is negligibly small compared with that of length; hence we may put with tolerable accuracy f(H)+2F(H)=0. With steel and soft iron, the change of volume is not very small compared with the change of length. But if t does not exceed 50 C. G. S. units, the effect of volume change on the change of length by combined action of t and t is negligibly small, for in these strong fields at which the change of volume is pronounced, the ratio  $t^2/H^2$  in the above expression becomes very small. Hence even for these metals, we may neglect the change of volume, provided the circular field is not very large, and the expression for  $\frac{\partial L}{L}$  becomes, in all cases,

$$\frac{\partial L}{L} = \frac{l^2 - \frac{1}{2} t_+^2}{II^2} t(II).$$

Since the material is supposed to be isotropic, f(H) is the same as the ordinary change of length by longitudinal magnetization. Thus the change of length by longitudinal magnetization with a constant circular field can be calculated from the change of length by longitudinal magnetization alone. The same expression can also be used for the calculation of the change of length due to circular magnetization with a constant longitudinal field.

In order to compare the above result with that of the experiment, it is obviously necessary to subtract from  $\frac{\partial L}{L}$  the expression F(t) for the change of length by longitudinal magnetization with a constant circular field t, and f(t) for the reciprocal case.

Assuming for the expression f(H) a suitable empirical formula for iron, steel or nickel, a simple analytical discussion of

the expression  $\frac{\partial L}{L}$ , or numerical calculation of it for different values of l and t from the experimental curve of the ordinary change of length leads to the conclusion that for iron, steel and nickel, all the points, which we have remarked in connection with the curves shown in Figs. 1, 4, 5, 7, 8 and 10, are involved without exception in the expression of  $\frac{\partial L}{\partial L}$ .

It may be observed that the behaviour of cobalt with regard to the change of length is just the reverse of that of iron, and therefore every result which we have obtained for iron is also applicable to the case of cobalt, provided the sign of the length change be properly reversed. Thus in strong fields, the length of a cobalt tube should, by the combined effect of longitudinal and circular magnetizations, become shorter than when acted upon by the former alone. In weak fields, the result should be just the opposite. The field of maximum contraction should increase with the circular, and the amount of the contraction, after reaching a maximum, gradually decrease. The circular field at which the maximum contraction occurs should be far greater than that for iron.

19. The comparison above made is qualitative; how the calculated and the experimental numbers agree with each other is seen from the following table:

98 K. Honda.

TABLE IX.

Nickel Tube, t=10.7							
l	L' (cal.)	L' (exp.)	difference				
10	$-20\times\overline{10}^{7}$	$-25\times10^{7}$	$5 \times \overline{10}^{7}$				
2)	- 74	- 85	11				
30	-112	-125	13				
50	<b>—</b> 162	-175	13				
80	-204	-216	12				
120	-237	-250	13				
200	-270	-285	15				
300	-291	<b>-</b> 305	14				
500	-309	-323	1-1				
700	-318	-331	13				

Here L' denotes  $\frac{\partial L}{L} - F(t)$ , and its value was calculated from the experimental curve for the ordinary change of length. A glance in the above table shows a fair agreement between the calculated and the experimental values. The difference between these numbers is not of a serious nature, if we remember that one scale division of the micrometer ocular corresponds to the change of  $5.12 \times 10^{-7}$  for nickel, and that the correction for temperature amounts to  $11 \times 10^{-7}$  in the most significant case.

The discrepancy is probably due to the residual effect and also to the coloropy of the tube. If the tube, after it is magnetized both longitudinally and circularly, is demagnetized by reversals with regard to the longitudinal magnetization, the circular field remaining constant, as was actually the case in the present experiment, the clongation due to the circular field alone is usually increased by one or two scale divisions,

a phenomenon which is perhaps to be attributed to the residual effect noticed in my former paper<sup>1)</sup>. The constancy of the difference in the above table furnishes additional evidence in support of this view. The colotropy of the tube as regards the change of length evidently influences the experimental values. Moreover the change of the intensity of longitudinal magnetization due to the mutual interaction of longitudinal and circular fields is not taken into account in the calculation of the effective field. These causes, I believe, are sufficient to account for the said discrepancy.

In steel and soft iron, there are comparatively large differences between the calculated and the experimental numbers, as will be seen from the following table:

,	Wolfram s	steel, $t=17.7$	Soft iron, t=26.2		
	L' (cal.)	L' (exp.)	L' (cal.)	L' (exp.)	
10	$2 \times 10^7$	$1 \times 10^{7}$	$9 \times 10^{7}$	$8 \times 10^{7}$	
30	22	18	23	30	
50	31	37	29	38	
80	39	51	31	41	
120	46	57	29	41	
200	48	62	21	36	
300	48	64	12	26	
500	47	63	- 5	7	
700	4.5	62	-17	- 5	

TABLE X.

For iron and steel, the sensibility of the apparatus was about  $2 \times 10^{-7}$  and the correction for temperature amounted to  $5 \times 10^{-7}$ 

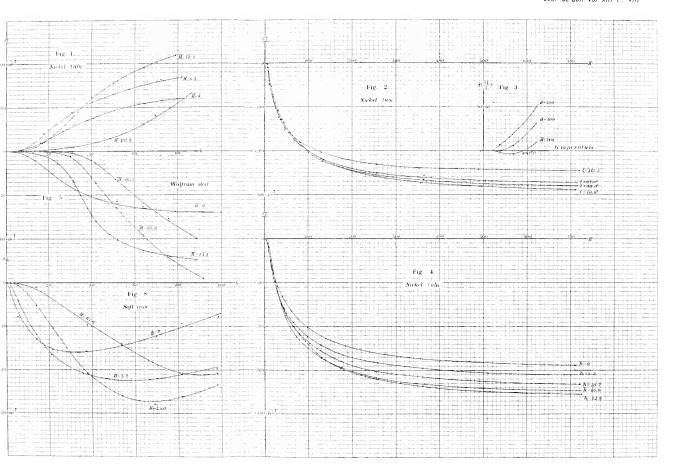
<sup>1)</sup> K. Honda, Jour. Sc. Coll. XI., 311, 1899.

in the most significant case. I believe that the principal causes of discrepancy above enumerated are sufficient to account for the difference between the calculated and the experimental numbers.

19. Thus qualitatively the above result and the experiment are in complete agreement with each other, although there are some discrepancies in quantitative details; there are, however, probable causes to account for the discrepancies. According to Knott, the change of length in cobalt by longitudinal magnetization is very little affected by the presence of a circular field, but the above consideration leads to a result which contradicts his anticipation. Hence a single experiment on this point for cobalt will decidedly establish the correctness of the one explanation against that of the other.

In conclusion, I wish to express my best thanks to Prof. H. Nagaoka, and also to Prof. A. Tanakadate for useful advice and kind guidance.





1(

in

oi

tl

n

aı

 $\mathbf{d}$ 

c٤

cl

li

e(

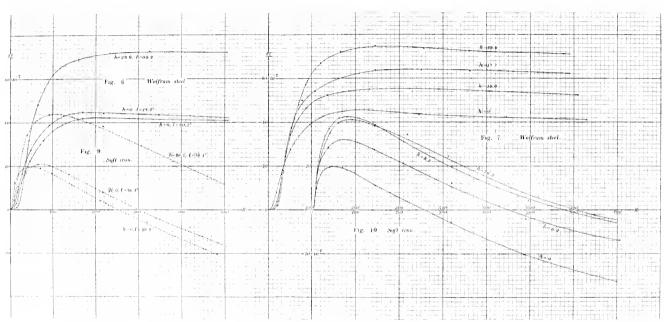
Н

de tl

Н

aı

.



				<u>.</u>	

# Studien über die Anpassungsfähigkeit einiger Infusorien an concentrirte Lösungen<sup>1</sup>).

Von

## Atsushi Yasuda, Rigakushi,

Professor der Naturgeschichte an der zweiten Hochschule zu Sendai.

Hierzu Tafel X-XII.

# Einleitung.

In der Natur finden wir Thiere und Pflanzen stets den obwaltenden Bedingungen angepasst. Diese Anpassung an die Umgebung ist in der That erst im Verlaufe langer Generationen hervorgebracht worden. Wenn dann aber die Lebensbedingungen sich ändern, wie werden diese Organismen dadurch beeinflusst? Unsere bisherigen Erfahrungen lehren uns, dass den Organismen im Allgemeinen die Fähigkeit innewohnt, sich diesen veränderten Verhältnissen genau anzupassen und so dauernd leben zu können, nur unter der Bedingung, dass die Veränderung nicht plötzlich stattfindet, oder, wenn sie rasch vor sich geht, sie doch verhältnissmässig unbedeutend ist.

<sup>&</sup>lt;sup>1</sup>) Die vorläufige Mittheilung dieser Arbeit erschien in The Botanical Magazine. Tokyo 1897. Vol. XI, No. 121. pp. 19-24. und Annotationes Zoologicae Japonenses. 1897. Vol. I, Part I et II. pp. 23-29.

Bekanntlich giebt es in der Pflanzenwelt die Wasserformen der amphibischen Gewächse, wie Polygonum amphibium und Ranunculus aquatilis, die sich in ihrer morphologischen und anatomischen Beschaffenheit ganz anders als ihre Landformen verhalten. Auch sind die Hydrophyten in Bezug auf Gestalt und Struktur einer grossen Metamorphose unterworfen, die sie zum Leben im Wasser befähigt. Aehnliche Thatsachen finden wir auch in der Thierwelt. Hierher gehören z. B. unter den Amphibien die Anuren, deren aus den Eiern ausschlüpfende Larven durch Kiemen athmen, aber im erwachsenen Zustande durch Lungen, während die Thiere, welche zu den Perennibranchiaten gehören, fortdauernd Kiemen besitzen, weil sie lebenslang im Wasser wohnen und niemals ein oberirdisches Leben führen, so dass sie sich jenem Medium völlig accommodirt haben.

Es dürfte daher von Interesse sein, wenn wir die Beschaffenheit der in der Natur gefundenen Medien verändern, künstliche Nährmedien anfertigen und die Anpassungsfähigkeit gewisser für diesen Zweck ausgewählter Organismen an diese künstlichen Medien studiren. Es liegen bereits Untersuchungen mancher Forscher über derartige Kulturversuche bei niederen Organismen vor. Im nächsten Abschnitt fasse ich die Resultate der wichtigsten einschlägigen Arbeiten zusammen.

Vorausschieken will ich noch, dass ich die Anpassung der Infusorien an solche künstlichen Medien studirt habe, die aus heterogenen Lösungen von höheren Concentrationen bestanden. Folgendes sind die Fragen, die ich zu beantworten versucht habe:—
1) Welche Grade der Concentrationen der Aussenmedien können die Infusorien ertragen? 2) Welche relative Widerstandsfähigkeit haben sie im Vergleich mit Algen und Pilzen? 3) Welche

Veränderungen ihrer äusseren und inneren Gestalt werden dadurch hervorgebracht, und wie wird ihr Bewegungs- und Vermehrungsvermögen durch diese heterogenen Medien beeinflusst? In Folgendem werde ich der Reihe nach die diesbezügliche Litteratur, die Methodik meiner Versuche, die Beschreibung der Experimente und endlich die Zusammenfassung der Resultate mittheilen.

Die vorliegende Arbeit wurde grossentheils im botanischen Institut der kaiserlichen Universität zu Tokyo unter freundlicher Anregung des Herrn Prof. Dr. M. Miyoshi ausgeführt, welchem ich für seine liebenswürdige Rathschläge meinen verbindlichsten Dank ausspreche. Herrn Prof. Dr. J. Matsumura sage ich auch an dieser Stelle für das Interesse, welches er meiner Arbeit entgegengebracht hat, meinen besten Dank.

#### Litteratur.

Ueber die Accommodation niederer Pflanzen giebt es ziemlich viele Versuche; so beobachtete Stahl¹), dass Aethalium septicum sich allmählich an Traubenzuckerlösungen anpasste und der Einwirkung einer 2%igen Lösung widerstand. Richter²) experimentirte mit Cyanophyceen und fand, dass Rivularia 3% ige, Gloeocapsa 6%ige, Anabaena 6%ige und Oscillaria 10%ige Kochsalzlösung ertragen konnten. Auch gelang es ihm, Diatomaceen in einer 7%igen Kochsalzlösung ein Jahr und in einer 10%igen einen Monat lang leben zu lassen. Er zog ausserdem

<sup>1)</sup> E. Stahl. Zur Biologie der Myxomyceten. Bot. Ztg. 1884. Nr. 11. p. 166.

<sup>&</sup>lt;sup>2</sup>) A. Richter. Ueber die Anpassung der Süsswasseralgen an Kochsalzlösungen. Flora. 1892. pp. 18-56.

verschiedene andere Algen in den Bereich seiner Untersuchungen. darunter Zygnema, Mougeotia, Spirogyra, Cosmarium, Chlorella, Tetraspora, Chaetophora, Vaucheria, Oedogonium, Chara u. s. w.; eine gewisse Anzahl von ihnen vermochte sogar in 13% iger Lösung zu existiren. Ferner theilte Klebs¹) mit, dass einige in den Lösungen organischer Verbindungen kultivirte Süsswasseralgen anfänglich eine Plasmolyse zeigten, die aber nach einigen Stunden vollständig ausgeglichen war, worauf sie ohne Schaden in den neuen Medien fortlebten. Nach demselben Autor gedieh Zugnema in einer 10 bis 20% igen Glycerinlösung eine Woche lang. Auch 10-50% ige Rohrzuckerlösungen vermochten dieselbe Alge im Leben halten, aber mit verschiedenen Wirkungen je nach der Concentration: 10% ige Lösung veranlasste lebhafte Kerntheilung, 20-25% ige Längenwachsthum, 30% ige Zellhautbildung und 40% ige Assimilation und Stärkebildung, während in 50% iger Lösung die Alge nur wenige Tage lebte.

Unter den Meeresalgen nahm Janse<sup>2</sup>) eine ähnliche Erscheinung bei Chaetomorpha wahr, und zwar hervorgebracht durch Kalisalpeter- und Kochsalzlösungen. Er fand nämlich, dass, wenn man diese Alge in eine solche Lösung legt, in Folge ihrer Anpassung an dieselbe nach kurzer Zeit ihre Widerstandsfähigkeit bedeutend gesteigert wird. Oltmanns<sup>3</sup>) machte Experimente über den Einfluss der Concentrationsänderung des Meerwassers auf Fucus, der bei niedriger Concentration sich dem neuen Medium gänzlich accommodirte. Eschenhagen<sup>4</sup>) kulti-

G. Klebs. Beiträge zur Physiologie der Pflanzenzelle. Berichte der deutsch. bot. Gesellsch. 1887. Bd. V, Heft 5. p. 181.

<sup>&</sup>lt;sup>2</sup>) J. M. Janse, Plasmolytische Versuche an Algen. Bot. Centralbl. 1887. Bd. XXXII. p. 21.

<sup>3)</sup> F. Oltmanns. Ueber die Bedeutung der Concentrationsünderung des Meerwassers für das Leben der Algen. Sitzb. d. Königl. preuss. Akad. d. Wissenseh, zu Berlin. 1891. p. 193.

<sup>4)</sup> F. Eschenhagen. Ueber den Einfluss von Lösungen verschiedener Concentration auf das Wachsthum von Schimmelpilzen. Stolp. 1889.

virte Aspergillus niger, Penicillium glaucum und Botrytis einerea in verschiedentlich concentrirten Lösungen von Traubenzucker, Glycerin, Natronsalpeter, Kalisalpeter, Chlornatrium und Chlorkalium, und wies sowohl die Grenzpunkte ihrer Accommodation als auch ihr Wachsthumsverhältniss zu diesen Substraten nach. Bachmann¹) bewies durch zahlreiche Experimente, dass Thamnidium elegans durch veränderte äussere Bedingungen gezwungen werden kann, diese oder jene Art von Sporangiolen zu bilden oder die Bildung derselben gänzlich zu unterdrücken. Ray²) säte die Sporen von Sterigmatocystis alba in Medien, welche aus Zucker, Stärke, Möhren, Kartoffeln, Gelatine und mineralischen Salzen bestanden, und erhielt verschiedene aus diesen Sporen entwickelte Pilzformen.

Auch für das Thierreich fehlt es an diesbezüglichen Untersuchungen nicht. Als Beispiele seien folgende angeführt:— Schmankewitsch<sup>3</sup>) beobachtete, dass Branchipus stagnalis, der immer in Süsswasser gefunden wird, sich, wenn man ihn in versüsstem Meerwasser züchtet, der Form von Artemia Milhausenii, einer das Brackwasser bewohnenden Art, nähert, und, wenn man das Brackwasser so lange concentrirt, bis dasselbe den Salzgehalt des Meerwassers erreicht hat, sich in Artemia salina, eine Meerwasser-Art, verwandelt. Herbst<sup>4</sup>) züchtete die Larven einiger Seeigel in verschiedenen Lösungen von Lithium-,

<sup>1)</sup> J. Bachmann. Einfluss der äusseren Bedingungen auf die Sporenbildung von *Thamnidium elegans* Link. Bot. Ztg. 1895. Abt. I. p. 128.

<sup>&</sup>lt;sup>2</sup>) M. J. Ray. Variations des Champignons inférieurs sous l'influence du milieu. Revue générale de Botanique. 1897. T. IX. pp. 193-259 et pp. 283-304.

<sup>3)</sup> W. Schmankewitsch. Zur Kenntniss des Einflusses der äusseren Lebensbedingungen auf die Organisation der Thiere. Zeitsch. f. wiss. Zool. 1887. Bd. XXIX. p. 429.

<sup>4)</sup> C. Herbst. Experimentelle Untersuchungen über den Einfluss der veränderten ehemischen Zusammensetzung des umgebenden Mediums auf die Entwicklung der Thiere, I. Theil. Zeitsch. f. wiss. Zool. 1892. Bd. XV. p. 446.

Natrium- und Kaliumsalzen, und fand, dass die Wirkungsstärke auf die Entwicklungsstufen derselben in einer Reihe der Salze von demselben Radical ihrem Molekulargewicht umgekehrt proportional ist, d. h. ihre Wirkungsstärke nimmt um so stärker ab, je mehr ihre Molekulargewichte zunehmen. Nach ihm wurde in einem Falle die Gastrulation der Seeigel bedeutend verzögert, in einem anderen Falle wurde die Pluteusorganisation entweder mit der runden und gedrungenen Gestalt ohne Fortsätze oder sogar ohne eine Spur des Kalkgerüstes gewonnen. Cohn¹) bemerkte, dass eine plötzliche Concentrationsänderung des Aussenmediums der Infusorien eine schädliche oder tödtliche Einwirkung ausübt. Fabre-Domergue<sup>2</sup>) beobachtete auch das Verhältniss der Ernährung in den Körpern einiger Infusorien, und gelangte zu folgendem Schluss: "Dans des conditions parfaites de nutrition prise dans l'acception la plus large du mot il se produit des aliments de réserve qui disparaissent quand des conditions deviennent défavorables à la vie." Weiter studirte Bokorny3) die Veränderungen der Bewegung, der Gestalt und der Grösse der Vacuolen von Paramaecium unter dem Einfluss gewisser Basen, wie Coffein, Ammoniak und Kali, deren 1 promill. oder noch dünnere Lösung im Allgemeinen die Bewegung verlangsamte, die Gestalt abrundete und sowohl Vergrösserung der Vacuolen als auch das Auftreten von neuen verursachte.

<sup>&</sup>lt;sup>1</sup>) F. Cohn. Entwickelungsgeschichte der microscopischen Algen und Pilze. Nova Acta Akad. Caes. Leopold. 1851. Bd. XXIV, Th. 1. p. 132.

<sup>&</sup>lt;sup>2</sup>) M. Fabre-Domergne. Recherches anatomiques et physiologiques sur les infusoires ciliés. Ann. d. Sc. nat. Zool. 1888. Sér. VII, T. 5. p. 135.

<sup>&</sup>lt;sup>3</sup>) Th. Bokorny. Einige vergleichende Versuche über das Verhalten von Pflanzen nd niederen Thieren gegen basische Stoffe. Pflüger's Archiv. 1895. pp. 557-562.

Bokorny gab auch in einer anderen Schrift (Vergleichende Studien über die Giftwirkung verschiedener chemischer Substanzen bei Algen und Infusorien. Pflüger's Archiv. 1896. pp. 262-306.) eine genaue Untersuchung über die Giftwirkung von Basen und Säuren unorganischer Natur, Salzen, Oxydations-Giften, Phosphor, organischen Säuren, Alkoholen, Alkaloiden u. a. m. auf das Leben der Infusorien und anderer Organismen.

Endlich müssen noch die Resultate der Untersuchungen von Davenport und Neal¹) Erwähnung finden; sie züchteten Stentor 2 Tage lang in einer 0,00005% Sublimat enthaltenden Kulturlösung; die Thiere liessen sich sehr wohl acclimatisiren und erwiesen sich gegen eine 0,001% ige sonst tödtliche Sublimatlösung ca. vier Mal länger widerstandsfähig als diejenigen, die im Wasser kultivirt worden waren.

Ueberblickt man die Ergebnisse dieser Untersuchungen, so ersieht man, dass sowohl den niederen Thieren als auch den niederen Pflanzen die Fähigkeit innewohnt, sich geänderten Aussenmedien leicht anzupassen. Da aber diese Fähigkeit bei verschiedenartigen Organismen verschieden stark ausgeprägt ist und unter Umständen mannigfaltig auftritt, so muss jeder specielle Fall genau erforscht werden. Meine vorliegenden Studien sollen in dieser Hinsicht einen kleinen Beitrag bringen.

#### Methodisches.

Als Versuchsmaterial wählte ich solche Infusorien aus, die in Grüben und Teichen stets gefunden werden können. Da aber die in der freien Natur vorkommenden Infusorien nie in reiner Kolonie vorhanden sind, so liess ich sie in einem Gefässe sich massenhaft entwickeln und unter Vorsichtsmassregeln eine längere Zeit fortleben.

Genau nach den Angaben von Miyoshi²) kultivirte ich die Infusorien in einem mit *Spirogyra* gefüllten Gefässe. Sobald

<sup>&</sup>lt;sup>1</sup>) C. B. Davenport and H. V. Neal. On the Acelimatization of Organisms to Poisonous Chemical Substances. Archiv für Entwicklungsmechanik der Organismen. 1895. Bd. II, Heft 4. p. 581.

<sup>&</sup>lt;sup>2</sup>) M. Miyoshi. Physiologische Studien über Ciliaten. The Botanical Magazine. Tokyo 1896. Vol. X, No. 112. p. 43.

die grüne Masse der Alge sich allmählich zu verfärben begann und die ursprünglich klare Flüssigkeit immer mehr getrübt wurde, bemerkte ich die Entwickelung verschiedener Arten von Infusorien, die sich oft mit einer erstaunlichen Schnelligkeit vermehrten und sich auffallenderweise in bald fadenförmigen, bald netzartigen Kolonien gruppirten. Die microscopische Untersuchung ergab, dass diese Kolonien nur aus wenigen Arten bestanden, die im Kampfe ums Dasein den Rest überwunden hatten. Aus dieser Mischkultur isolirte ich einzelne Arten, indem ich mittelst einer Pipette eine kleine Menge der Kolonie zusammen mit Wasser herausholte und in ein ebenfalls mit Brunnenwasser und der Alge gefülltes Gefäss versetzte. Bei gewöhnlicher Zimmertemperatur zeigten diese Kulturen ca. in einer Woche üppige Entwickelung; nach vier oder fünf Wochen aber nahm die Vermehrung ab, und endlich nach sechs Wochen konnte nur noch eine ausserordentlich kleine Anzahl in der Flüssigkeit gefunden werden. Um eine und dieselbe Art immer in üppiger Kultur zu halten, legte ich deshalb alle drei Wochen neue Kulturen an und trug dafür Sorge, dass sie nicht etwa durch Bacterien inficirt wurden.

Nachdem ich die gewünschten Arten auf solche Weise in Kultur hatte, wurden die Experimente auf zweierlei Weise ausgeführt: einerseits prüfte ich die Anpassungsfähigkeit der Infusorien in dem Zustande, wie sie in der Natur vorkommen, d. h. in ihrem Zusammenleben mit Bacterien; anderseits wandte ich zu demselben Zwecke die Reinkultur jedes Infusors an, also frei von Bacterien.

Der grösste Theil meiner Experimente wurde mit unreinen Kulturen ausgeführt; in einigen Fällen wiederholte ich die Experimente an Reinkulturen, um zu wissen, ob die Gegenwart der Bacterien etwa das Ergebniss der Experimente modificirt hätte. Die Resultate stimmten aber bei beiden Kulturen vollkommen überein, wie wir nachher sehen werden.

Als äussere Medien verwendete ich Lösungen von Rohrzucker, Traubenzucker, Milchzucker, Glycerin, Kalisalpeter, Natronsalpeter, Chlorkalium, Chlornatrium und Chlorammonium in verschiedenen Concentrationen. Diese Stoffe waren chemisch rein und wurden vor dem Gebrauch vollständig getrocknet. Folgende Infusorien wurden bei meinen Studien ausschliesslich verwendet: Euglena viridis, Chilomonas paramaecium, Mallomonas Plosslii, Colpidium colpoda und Paramaecium caudatum. Alle Kulturen, sowohl unreine als reine, wurden bei Zimmertemperaturen von 25°-30° C gehalten und in den Wintermonaten in einen Thermostat von etwa 30° C gestellt.

Für unreine Kulturen benutzte ich eine grosse Anzahl der 5 cm hohen und 3 cm weiten cylindrischen Glasgefässe, deren jedes 30 ccm der Versuchslösung und etwa 1 Gramm Spirogyrα-Fäden enthielt. Da die Infusorien im Brunnenwasser weit besser gedeihen als in destillirtem Wasser, so wendete ich bei der Zubereitung der flüssigen Versuchsmedien das letztere als auflösendes Mittel für verschiedene Substanzen an, wobei die Menge der darin gelöst vorhandenen Stoffe mit Ausnahme von Kochsalz¹) so unbedeutend war, dass ich sie ohne grosse Ungenauigkeit ausser Acht lassen konnte. Bei vielen Kulturen, die gleichzeitig gemacht wurden, nahm ich keinen Anstand, eine Kontrollkultur, in welcher nur Brunnenwasser und Spirogyrα-Fäden angewendet wurden, anzufertigen und zum Vergleiche dienen zu lassen.

In Bezug auf die Reinkultur der Protozoen im Allgemeinen

 $<sup>^{\</sup>mbox{\tiny 1}})$  Die chemische Analyse des Brunnenwassers zeigte, dass es 0,095% Kochsalz enthielt.

haben viele Forscher¹) in neuerer Zeit versucht, sie entweder auf festen Substraten oder in flüssigen Medien zu züchten. Bei der Isolirung der Infusorien befolgte ich genau die Methode von Ogata²) mit positivem Resultat. Ich liess mir nach seiner Vorschrift feine Glascapillarröhren anfertigen, deren Durchmesser je nach der Grösse des Versuchsobjects variirten. So war z. B. bei Chilomonas paramaecium, dessen Körper 25-30 μ Länge und 10-12 μ Breite hat, das Capillarrohr etwa 0.1 mm in innerem Durchmesser und 10 cm in Länge, während es bei Calpidium colpoda, dessen Körper 60-70 μ lang und 25-30 μ breit ist, einen inneren Durchmesser von 0,15 mm und eine Länge von 10 cm hatte.

Sobald das Capillarrohr nach dem Eintauchen in eine sterilisirte Nährlösung mit der letzteren grossentheils gefüllt war, brachte ich das nämliche Ende desselben in die Mischkulturflüssigkeit von Bacterien und Infusorien und liess das Rohr sich mit der Flüssigkeit völlig füllen. Untersucht man ein solches Capillarrohr unter einem Microscope, so findet man an der Capillarrohrmündung eine grosse Anzahl von Infusorien in Bewegung. Einige streben sich ins Innere zurückzuziehen, bald aber kommen sie nach der Mündung zurück. Wegen der starken Aërotaxis und schwachen Chemotaxis der Organismen<sup>3</sup>) gelingt

<sup>1)</sup> Während Beijerinck (Kulturversuche mit Amöben auf festen Substraten. Centralbl. f. Bak. u. Parasit. 1896. Bd. XIX, No. 8), Celli (Die Kultur der Amöben auf festen Substraten. Centralbl. f. Bak. u. Parasit. 1896. Bd. XIX, No. 14/15.), Schardinger (Reinkulturen von Protozoen auf festen Nährboden. Centralbl. f. Bak. u. Parasit. 1896. Bd. XIX, No. 14/15.), Gorini (Die Kultur der Amöben auf festen Substraten. Centralbl. f. Bak. u. Parasit. 1896. Bd. XIX, No. 20), Tischutkin (Ueber Agar-Agarkulturen einiger Algen und Amöben. Centralbl. f. Bak., Parasit. u. Infekt. 1897. Bd. III, No. 7/8.) und andere Forscher Amöben auf festen Substraten künstlich züchten konnten, ist es Ogata (Ueber die Reinkultur gewisser Protozoen-Infusorien. Centralbl. f. Bak. u. Parasit. 1893. Bd. XIV, No. 6.) auch gelungen, Polytoma wella in flüssigen Medien rein zu kultiviren.

<sup>2)</sup> M. Ogata. loc. cit. p. 168.

<sup>3)</sup> M. Miyoshi, loc. cit. p. 48.

es nicht immer, die Infusorien auf diese Weise hervorzulocken, und ihrer habhaft zu werden. Nur wenn sie zufällig in die sterilisirte Flüssigkeit tief eindringen, kann man unter dem Microscope das Capillarrohr an der betreffenden Stelle abbrechen und dann das Ende des Rohrs zusehmelzen. Sodann impft man den infusoriumhaltigen Capillarrohrinhalt, indem man das Rohr mit einem sterilisirten Pincet abbricht und den Inhalt in ein mit sterisirter Nährlösung gefülltes Reagensglas hineinbläst.

Bei meinen Versuchen impfte ich wenigstens zwei Individuen in ein und dasselbe Reagensglas, um erstens den Effect der Inoculation zu siehern, und zweitens mit der Hoffnung, dass sie, wenn alle beide in dem neuen Medium unversehrt fortlebten, durch Copulation sich vermehren könnten. Bei der Zimmertemperatur von 20° C blieb die geimpfte Nährflüssigkeit nach zwei oder drei Tagen vollkommen klar, und erst nach ungefähr zehn Tagen erschienen Hunderte von Individuen, die nahe der Oberfläche der Flüssigkeit als sehr kleine weisse Pünktchen hin und her schwammen. Die Zahl dieser weissen Pünktchen nahm hernach allmählich zu, und dieselben waren nicht allein am oberen Theil des Reagensglases, sondern auch am mittleren und unteren Theil desselben zerstreut sichtbar. Eine solche Erscheinung bedeutet, dass die Reinkultur gut ausgefallen ist, und dass man in jener Nährlösung nichts anders als die isolirte Art der Infusorien findet. Wenn aber die Nährlösung während der Impfung von Bacterien inficirt wird, so tritt immer eine starke Trübung zu Tage, und man bekommt in diesem Falle selbstverständlich keine Reinkultur der Infusorien.

Die auf diese Weise hergestellte Reinkultur gedieh 4-5 Wochen lang, vorausgesetzt dass die Nährstoffe in der Kultur-flüssigkeit nicht völlig erschöpft waren. Durch erneuerte Wieder-

impfung konnte ich die Organismen in reinem, gutem Kulturzustande eine lange Zeit erhalten.

Die Nährlösung, die ich für die Reinkultur gebrauchte, stellte ich nach der Vorschrift von Ogata an, und zwar war ihre Zusammensetzung folgende:

Fleischextract	<b>1</b> g	
Rohrzucker	20 ,,	
Concentrirt gekochte Lösung von Porphyra vulgaris.	$250 \mathrm{ccm}$	1
Destillirtes Wasser	729 ,,	

### Beschreibung der Versuche.

Wie gesagt, stellte ich die Experimente hauptsächlich mit unreinen Kulturen an, in der Absicht, den Einfluss, welchen die äusseren Bedingungen auf die Infusorien in ihrem natürlichen Vorkommen ausüben, festzustellen. Dabei versäumte ich aber nicht, Kontrollversuche mit reinen Kulturen zu machen und die beiden Resultate zu vergleichen.

Bei allen Versuchen mit unreinen und reinen Kulturen liess ich die Beschaffenheit des Mediums sich plötzlich ändern und prüfte die Anpassungsfähigkeit unserer Organismen an das neue Medium. Hatte ich unreine Kulturen, so verglich ich gewöhnlich im Verlauf von 1-7 Tagen, zuweilen aber erst nach einem Monat, die Wirkungen der verschiedenen Mediumsconcentrationen auf das Reproductionsvermögen und die Gestaltänderungen der Versuchsorganismen. Speciell bei den Versuchen mit Rohrzucker war es nöthig, eine Reinkultur anzuwenden, weil bei unreiner Kultur der Rohrzucker durch vorhandene Bacterien oder Pilze nach und nach invertirt und schliesslich gespalten wurde. Um

zu erkennen, nach wie vielen Tagen der Rohrzucker zum Traubenzucker invertirt wird, prüfte ich mit der Fehling'schen Lösung und fand, dass in meinen Versuchen nach etwa 4 Tagen eine kleine Inversion stattgefunden hatte. Meine unreinen Rohrzuckerkulturen waren daher binnen der ersten drei Tage doch noch brauchbar.

Dass diese Inversion ausser durch Bacterien und Pilze auch unter Mitwirkung der Infusorien stattfände, ist schon von vorn herein unwahrscheinlich. Um dies aber experimentell zu constatiren, stellte ich einige Versuche mit Rohrzuckerreinkulturen¹) an, und gelangte beim Prüfen der fraglichen Flüssigkeit wie erwartet zu negativem Resultat. Die Kontrollkultur mit Aspergillus glaucus zeigte eine starke Inversion.

Bei allen Kulturen mit verschiedenen Stoffen machte ich immer Kontrollkulturen, und bei den kritischen Versuchen, wie z. B. der Bestimmung der Concentrationsgrenze einer Flüssigkeit, in welcher die Organismen sich mehr oder minder anpassend leben können, wurden dieselbe Kulturen einige Male wiederholt.

Ich gehe nun zur Beschreibung der einzelnen Versuche bei jeder Art meiner Versuchsorganismen über.

# (a) Euglena viridis Ehrbg.<sup>2</sup>)

Dieser Organismus hatte in der Kontrollkultur folgende

<sup>&</sup>lt;sup>1</sup>) Da der Rohrzucker bekanntlich bei langem Kochen zum Theil in Trauben- und Fruchtzucker verwandelt wird, so kann bei den Rohrzuckerreinkulturen die gebräuchliche Sterilisirung durch Hitze nicht ohne Vorsichtsmassregeln angewendet werden. Ich sterilisirte deshalb den Rohrzucker mit absolutem Alkohol und brachte ihn dann in die vorher sterilisierte Nährlösung ein.

<sup>&</sup>lt;sup>2</sup>) Figuren in Friedrich Ritter v. Stein, Der Organismus der Infusionsthiere. Leipzig 1878. Abt. III, Heft I. Taf. XX., W. Saville Kent, A Manual of the Infusoria. 1880-81. Vol. I. Pl. XX. und O. Bütschli, H. G. Bronn's Klassen und Ordnungen des Thierreiches. 1883-87. Bd. I. Protozoa, Abt. II. Taf. XLVII.

Merkmale: Gestalt gewöhnlich spindelförmig, Hinterende schärfer zugespitzt, aber wegen der Metabolie sich mannigfaltig verändernd. Aus dem Schlunde entspringt eine lange Geissel. Chromatophoren zahlreich vorhanden, klein, scheibenförmig und rein grün gefärbt. Eine contractile Vacuole nahe dem Vorderende gelegen. Dicht bei demselben Ende befindet sich auch ein rother Augenfleck.

Versuch 1. Rohrzucker, C12H22O11.—Von einer 1% igen Lösung anfangend liess ich in anderen Kulturen die Concentration um je 1% steigen. Obgleich die Accommodation schwer wurde, als die Concentration zunahm, so lebte das Infusor doeh bis zur 15%igen Lösung, welche die Maximaleoncentration für den Organismus war. 1% ige, 2% ige und 3% ige Kulturen zeigten keine wesentliche Veränderung am Körper des Organismus. Bei einer 4% igen Lösung aber begannen die Chromatophoren an Grösse zuzunehmen. Von 1% iger Lösung bis zu 7% iger war die spirale Bewegung des Organismus lebhaft, dagegen über 8% wurde sie allmählich langsamer, während die Chromatophoren selbst sich merklich ausdehnten; als die Concentration des Mediums zunahm, wurde auch die Vermehrung verhindert. Bei 12%iger Lösung konnte das Thier nicht mehr normal gedeihen, bei 13% überlebte eine kleine Anzahl, die jedoch nach einer Woche alle zu Grunde gingen; bei 14% lebten noch einige Individuen, aber nicht länger als 4 Tage, während sie bei 15% kaum einen Tag lebendig blieben. Da der Organismus metabolisch ist, so konnte keine deutliche Veränderung an seiner äusseren Gestalt beobachtet werden.

Versuch 2. Traubenzucker, C<sub>6</sub>H<sub>12</sub>O<sub>6</sub>.—Unser Organismus konnte 1-11% ige Concentrationen ertragen. Bei 1% - und 2% - Kultur war noch keine merkliche Veränderung wahrzunehmen,

aber schon bei 3% iger Lösung dehnten sich die Chromatophoren ein wenig aus, und bei der Concentration über 3% wurden sie noch etwas grösser. Die Bewegung des Thierkörpers schien bei 1-6%-Kulturen normal zu verlaufen; erst bei 7% wurde sie langsamer mit gleichzeitiger Verminderung der Vermehrungsfähigkeit. Bei einer 9% igen Lösung vermehrten sich die Thiere überhaupt nicht mehr, und nach einer Woche war nur noch eine kleine Anzahl am Leben. Alle Individuen des Infusoriums gingen bei einer 10%- Kultur nach einer Woche, und bei einer von 11% schon nach einigen Tagen zu Grunde.

Versuch 3. Milchzucker, C<sub>12</sub>H<sub>22</sub>O<sub>11</sub>+H<sub>2</sub>O.—Unter den oben erwähnten Zuckerarten schien unser Infusor sich an Milchzucker am besten anzupassen. Die Maximalconcentration, welcher es widerstehen konnte, war eine 17%ige. Von 4% an aufwärts schienen die Chromatophoren sich zu vergrössern. Bei 1-11%-Kulturen nahm die Multiplication rasch zu, aber über 12% wurde sie etwas vermindert und auch die Bewegung wurde einigermassen träge. Eine 17%ige Lösung erwies sich als die Grenzeoncentration für das Versuchsthier.

Versuch 4. Glycerin, C<sub>3</sub>H<sub>8</sub>O<sub>3</sub>.—Die Versuche lehrten uns, dass sich unser Infusor an Glycerin weit schlechter anpasste als an eine der oben erwähnten drei Zuckerarten, denn das Thier konnte nur 1-6% ige Lösungen ertragen. Bei einer 2% igen Lösung erweiterten sich die Chromatophoren; bei 3%-Kultur lebte eine kleine Anzahl noch am fünften Tage, und bei 6% blieben nur wenige Individuen noch einige Tage am Leben. Die Bewegung wurde bei einer 4% igen Concentration schon vielfach retardirt, und bei 6% hörte sie fast gänzlich auf. Ferner war in der letzteren Lösung eine pathologische Erscheinung wahrzunehmen, indem die Cuticula des Körpers um die

Chromatophoren etwas einschrumpfte, sodass ihr Umriss im optischen Schnitte gesehen zickzackförmig aussah, und die Chromatophoren selbst verschmolzen mehr oder weniger mit einander.

Versuch 5. Schwefelsaures Magnesium, MgSO<sub>4</sub>.—Unter den unorganischen Substanzen erwies sich das schwefelsaure Magnesium als dem Leben des Organismus am besten zusagend. Der Organismus konnte die Concentration von 1-6% vertragen. Die Chromatophoren nahmen in ihrer Grösse fortwährend zu, als die Concentration von 1,5% bis auf ihr Maximum stieg. In einer 3,4% igen Lösung zeigte das Thier eine sehr träge Bewegung, und schon bei 4-6% igen Lösungen ging es beinahe zum Stillstand über. Betrug die Concentration nur 1-2,5%, so gedieh unser Thier einen Monat lang vollkommen normal, aber von einer 2,6% igen Concentration an aufwärts büsste es seine Vermehrungsfähigkeit ein, und endlich bei 5-6% blieben nur vereinzelte Individuen am Leben.

Versuch 6. Salpetersaures Kalium, KNO<sub>3</sub>.—Der Organismus widerstand einer 2,4% igen Concentration. Von 0,8% an fingen die Chromatophoren an sich zu erweitern; über 2% wurde die Bewegung sehr langsam. Im Allgemeinen schien das vorliegende Salz auf die Mulptiplication des Infusors hemmend zu wirken, da selbst bei verdünnten Lösungen das Thier eine unbedeutende Vermehrung zeigte.

Versuch 7. Salpetersaures Natrium, NaNO<sub>3</sub>.—Dieses Salz verhielt sich fast wie das vorige. Eine 2% ige Lösung war das Maximum, welches unser Thier ertragen konnte. Die Chromatophoren dehnten sich schon von 0,8% an aus. Die Bewegung war bei 2% iger Lösung nach zwei Tagen sehr träge.

Versuch 8. Chlorkalium, KCl.—Nächst dem Magnesium-

sulfat erwies sich unter den unorganischen Stoffen das Chlorkalium für die Vermehrung des Organismus am günstigsten. Eine 0,7%ige Lösung verursachte sowohl Zahlvermehrung als auch Volumenerweiterung der Chromatophoren. In 0,2-1%igen Concentrationen gedieh der Organismus noch nach 40 Tagen. Stieg die Concentration auf 2,8%, welches die maximale Grenze für den Organismus war, so hörte die Bewegung fast gänzlich auf, während die Chromatophoren sich theilweise zu grösseren Körnern verschmolzen.

Versuch 9. Chlornatrium, NaCl.—0,2-1,8% waren die Concentrationen, bei welchen das Thier am Leben blieb. Die Chromatophoren schienen bei einer 0,8% igen Lösung an Grösse zuzunehmen, und bei einer 1,6% igen Concentration zeigte der Organismus noch eine langsame Bewegung.

Versuch 10. Chlorammonium, NH<sub>4</sub>Cl.—Dieses Salz wirkte unter allen oben genannten Stoffen am ungünstigsten auf das Leben des Organismus ein, sodass die Anpassungsgrenze hier am niedrigsten war. 0,2-0,6% Kulturen gediehen noch am Ende der dritten Woche, aber über 1% vermehrte sich das Thier nicht mehr, und bei 1,4% lebten kaum noch einige Individuen. Bei 0,6% iger Lösung nahm die Grösse der Chromatophoren zu, und bei 1% verschmolzen sie sich zu wenigen grösseren Körnern. Eine 1,4% ige Lösung verursachte immer die Verschmelzung der Chromatophoren und hob gleichzeitig die Bewegung des Organismus auf.

Ich wiederholte dieselbe Versuche zehnmal mit Reinkulturen und verglich die Resultate mit denjenigen bei den unreinen Kulturen. Die Ergebnisse stimmten in beiden Fällen völlig überein. Weiter beobachtete ich, dass bei den Versuchen mit Reinkulturen die Schnelligkeit der Multiplication für die Lösungen verschiede-

ner Concentrationen eines und desselben Stoffes nicht allzu gleich war, obgleich sie gleichzeitig geimpft worden waren Im Allgemeinen nahm die Vermehrungsenergie in dem Masse ab, wie die Concentration des Mediums stieg; so war zum Beispiel bei einer Traubenzuckerkultur, im Verlaufe von 2 Wochen nach der Impfung, die Vermehrung eine starke bei 2%, eine mässige bei 4%, eine sehr unbedeutende bei 6%, eine noch spärlichere bei 8% und keine bei 10%. Ferner war die Vermehrung bei derselben Kultur nach 4 Wochen bei 2% eine sehr starke, bei 4% eine starke, bei 6% eine mässige, bei 8% eine unbedeutende und bei 10% eine höchst spärliche Vermehrung zu beobachten, während sich am Ende der sechsten Woche bei 2-4% eine sehr starke, bei 6% eine starke, bei 8% eine mässige und bei 10% eine spärliche Vermehrung zeigte. Auch beim Milchzucker wurden ähnliche Thatsachen constatirt. So gediehen nach 2 Wochen 2-4%-Kulturen ausgezeichnet; 6%-Kultur zeigte eine starke, 8% eine mässige, 10% eine spärliche, 12% eine noch schwächere und 14% gar keine Multiplication mehr. Nach 4 Wochen aber vermehrten sich die Organismen bei 2-6% sehr stark, bei 8% stark, bei 10% mässig, bei 12% spärlich und bei 14% sehr spärlich. Endlich nach 6 Wochen gedieh die Multiplication stark bei 10%, mässig bei 12% und spärlich bei 14%. Auch für schwefelsaures Magnesium, salpetersaures Kalium, Chlornatrium u. s. w. habe ich ähnliche Erscheinungen wahrgenommen.

# (b) Chilomonas paramaecium Ehrbg.<sup>1</sup>)

Der Organismus in Kontrollkultur hatte folgende Charakteristika: Körper nicht metabolisch, sondern plastisch. Gestalt

<sup>1)</sup> Figuren in Friedrich Ritter v. Stein, loc. cit. Taf. XIX, W. Saville Kent. loc. cit. Pl. XXIV und O. Bütschli, loc. cit. Taf. XLV.

länglich oval, seitlich comprimirt. Vorderende breiter und schief abgestutzt, Hinterende dagegen rundlich zugespitzt. Zwei Geisseln am Vorderende, eine derselben rollte sich, wenn der Organismus im Ruhezustande war. Zahlreiche sphäroidische Amylumkörner dicht unter der Körperoberfläche. Eine contractile Vacuole am Vordertheil des Körpers.

Versuch 1. Rohrzucker.—Mit einer 1% igen Lösung anfangend liess ich die Concentration um je 1% steigen, wie es bei Euglena viridis der Fall war. Von 2% an aufwärts dehnten sich die Körnchen fortwährend aus. Ueber 6% nahm der Organismus an Dicke und Breite zu, nicht aber an Länge, und sah so einfach oval aus. Die Vermehrung wurde schon bei 4% verzögert; bei 7% lebten einige Individuen noch eine Woche lang. Die Individuen aus der 7% igen Kultur waren so träge, dass sie an einem bestimmten Platze still lagen und nur eine zitternde Bewegung zeigten; vor ihrem Tode hüpften sie einige Male rückwärts. Die letztere Erscheinung wurde auch bei den concentrirten Lösungen anderer Stoffe beobachtet.

Versuch 2. Traubenzucker.—Der Traubenzucker wirkte stärker als der Rohrzucker. Die höchste Concentration, die der Organismus vertragen konnte, war 6%. Bei 4%-Kultur ging die Vermehrung nicht mehr gut vor sich, und bei 5% blieb nur eine kleine Anzahl der Individuen am Leben. 2%ige Concentration bewirkte, dass die Körnchen sich vergrösserten, und bei 5% wurde die Bewegung sehr langsam. Bei 6% kam der Organismus fast zum Stillstande, und wurde eine Unebenheit des Körperumrisses hervorgerufen.

Versuch 3. *Milchzucker*.—Der Organismus widerstand 1-8%-igen Concentrationen. Ueber 3% vergrösserten die Körnehen ihr Volumen, bei 6% wurde die Multiplication verhindert und end-

lich bei 8% lebten nur noch einige Individuen eine Woche lang weiter, mit dem erwähnten Unebenwerden der Körperumrisse. Merkwürdig war, dass der Körper an Dicke und Breite zunahm, als die Concentration stieg.

Versuch 4. Glycerin.—Eine 4% ige Lösung war die Maximalconcentration für die Accommodation des Thieres. Bei 2% erweiterten sich die Körnchen, bei 3% hörte die Vermehrung auf, und bei 4% lebten nur noch einige Individuen, deren Körper unregelmässige Umrisse zeigten.

Versuch 5. Schwefelsaures Magnesium.—In 1-3% igen Lösungen lebte der Organismus fort. Von 0,8% an aufwärts vergrösserten sich die Körnchen, und in 3% iger Lösung wurden dieselben auffallend gross. Eine 1,4% ige Lösung verhinderte die Multiplication. Bei höheren Concentrationen trat bei einigen Individuen ein unregelmässiges Aussehen zu Tage. Diese Gestaltänderung wurde bei einer 2,5%-Kultur besonders gut beobachtet, indem alle Individuen noch 2 Wochen mit einer ungewöhnlichen Unebenheit ihrer Körpergestalt fortlebten.

Versuch 6. Salpetersaures Kalium.—Eine 2% ige Concentration schien die obere Grenze der Anpassung zu sein. Eine 0,8% ige Lösung verunlasste eine Vergrösserung der Amylumkörner, die bei einer 1% igen Lösung nach einer Woche einen sehr grossen Durchmesser zeigten. Die Vermehrung ging nur bei niederen Concentrationen gut vor sich.

Versuch 7. Salpetersaures Natrium.—Der Organismus konnte in 0,2-1,2% igen Lösungen leben. Die Volumenzunahme der Körnchen fand von 0,6% an statt. Bei höheren Concentrationen gedieh unser Organismus nicht. Im Uebrigen fast dieselben Erscheinungen wie beim vorhergehenden Versuche.

Versuch 8. Chlorkalium.—Eine 0,8%-Kultur am Ende des

dritten Tages nach der Impfung untersucht zeigte sowohl Vergrösserung der Körnchen als auch Abrundung des Körpers. Nach Verlauf einer Woche besassen einige Individuen in einer 1% igen Lösung eine fast scheibenförmige Gestalt. Ueber 2% konnten sie nicht mehr leben.

Versuch 9. Chlornatrium.—Eine 0,4% ige Lösung liess die Körnchen sich erweitern. Bei 0,8-1%-Kulturen dehnte sich ihr Volumen bedeutend aus, indessen der Körper des Organismus sich verkürzte und rundlich wurde. Bei 1% iger Concentration war die äusserste Grenze der Accommodation erreicht.

Versuch 10. Chlorammonium.—Der Organismus konnte sich an 0,1-0,6% ige Concentrationen anpassen. Bei einer 0,2% igen Lösung trat schon Körnehenvergrösserung ein, und bei 0,4% zeigten einige Individuen unebene Umrisse, mit gleichzeitiger Abschwächung ihrer Bewegung. Wie bei Euglena viridis so auch bei Chilomonas paramaecium übte der vorliegende Stoff von allen augewandten Chemikalien die stärkste Einwirkung aus.

# (e) Mallomonas Plesslii Perty.¹)

Der Organismus in der normalen Kultur zeigte folgende Merkmale: Gestalt oval, am Vorderende etwas schmaler. Die ganze Cuticularoberfläche mit langen, biegsamen, borstigen Wimpern bekleidet; am Hinterende mit einer langen Geissel versehen. Anstatt der Amylumkörner war eine Anzahl von Vacuolen vorhanden. Eine contractile Vacuole befand sich nahe dem hinteren Ende. Das Thier schwamm mit lebhafter Bewegung, wobei es oft plötzlich stillstand.

Versuch 1. Rohrzucker.—Der Organismus vertrug Anpas-

<sup>1)</sup> Figuren in W. S. Kent. loc. cit. Pl. XXIV.

sungsconcentrationen von 1-7%. Auch bei höheren Concentrationen nahm die Grösse des Körpers mehr oder minder zu, während die Multiplication und die Lebhaftigkeit der Bewegung allmählich sanken. Ferner wuchs die Zahl und Grösse der Vacuolen mit der Concentrationserhöhung bedeutend an; diese Erscheinung trat schon bei einer 2%-Kultur ein. Erst von 4% an wurde die Multiplication schwächer und bei 7% erlitt die Bewegung eine Retardirung, welche zu der sehr schnellen, normalen Bewegung in grossem Contrast stand.

Versuch 2. Traubenzucker.—Eine 6%-Kultur war das Maximum der Anpassung des Organismus. Die Vergrösserung des Körpers und die Zunahme der Vacuolen bei stärkeren Concentrationen waren wie beim Rohrzucker. Eine 2%ige Lösung erweiterte die Vacuolen einigermassen, und von 3% an nahm die Vermehrung des Organismus ab. Die Bewegungshemmung war schon bei 4% zu beobachten, noch stärker bei 5%.

Versuch 3. Milchzucker.—Das Infusor ertrug 1-9% ige Lösungen. Das allgemeine Resultat stimmte mit dem bei den anderen Zuckerarten überein; der einzige Unterschied war der, dass der Milchzucker eine schwächere Einwirkung auf den Organismus ausübte als die anderen Zuckerarten. Die Vacuolen fingen erst bei einer 3% igen Lösung an sich zu vermehren, und die Multiplication wurde erst von 7% an etwas verlangsamt.

Versuch 4. Glycerin.—Die Grenze der Accommodation war eine 4% ige Lösung. Bei einer 2%- und noch auffallender bei einer 3%-Kultur fand Zunahme der Zahl und Grösse der Vacuolen und Anschwellen des Organismus statt.

Versuch 5. Schwefelsaures Magnesium.—Der Organismus vermochte sich 1-3,4% igen Lösungen anzupassen. Eine 0,8% ige Concentration verursachte sowohl Vermehrung als auch

Vergrösserung der Vacuolen, und bei höher concentrirten Lösungen war Verhinderung der Multiplication und Retardation der Bewegung wahrzunehmen.

Versuch 6. Salpetersaures Kalium.—Eine 0,7%ige Lösung vergrösserte die Vacuolen etwas. Die Bewegung begann bei 1-1,5%igen Lösungen sehr langsam zu werden. Bei den Kulturen höherer Concentrationen pflegte der Organismus sich nicht zu vermehren, und im Verlaufe einiger Tage ging der grössere Theil der Individuen zu Grunde. Eine 1,5%ige Lösung bildete die Grenze der Anpassung.

Versuch 7. Salpetersaures Natrium.—Für diesen Stoff besass der Organismus eine besonders grosse Resistenzkraft. Er vermochte sich sogar einer 2,6% igen Concentration anzupassen, wenn auch mit grosser Schwierigkeit. Die Bewegung war bei 1,5% noch lebhaft.

Versuch 8. Chlorkalium.—Bei einer 0,8%-Kultur vergrösserten sich die Vacuolen. Die Grenze der Anpassung des Organismus war bei 1,4% iger Lösung zu beobachten. Mit der Concentrationssteigerung trat Körperabrundung ein.

Versuch 9. Chlornatrium.—Die Maximalconcentration war 1,5%. Bei 0,8%-Lösung sehien der Körper nach 5 Tagen sich abzurunden. In einer 1% igen Lösung konnte das Thier 3 Wochen lang gedeihen, aber in 1,5% starb es schon am Ende des vierten Tages.

Versuch 10. Chlorammonium.—Für diesen Stoff besass der Organismus die kleinste Anpassungsfähigkeit, ganz wie es bei den anderen Infusorien der Fall war. Eine 0,8% ige Lösung war das Maximum. Die Cuticularoberfläche des lebenden Organismus zeigte in dieser Lösung nach einem Tage einige longitudinale Falten.

# (d) Colpidium colpoda Ehrbg.<sup>1</sup>)

Merkmale des Organismus in der normalen Kultur:— Körper mittelgross, nierenförmig. Rückenseite mässig gewölbt; Bauchseite in der Nähe des Mundes etwas eingebuchtet. Vorderende viel schmaler als das abgerundete Hinterende. Cuticularwimpern auf der ganzen Oberfläche des Körpers reichlich vorhanden und an Grösse alle gleich. Mund in mässiger Entfernung vom Vorderende, in einer die Bauchseite querenden Einbuchtung. Eine contractile Vacuole und einige Nahrungsvacuolen vorhanden. Bewegung lebhaft.

Versuch 1. Rohrzucker.—Die Concentrationsdifferenz der Versuchsserie war 1%. 8% wurde als das Maximum erkannt. Schon bei einer 3%igen Lösung begannen die Vacuolen sich etwas zu vermehren und zu vergrössern. Diese Erscheinung wurde mit der Concentrationserhöhung immer mehr merklich. Ueber 4% sah der Körperumriss rundlich aus und die Grösse nahm merkwürdig zu. Multiplicationshemmung sehon bei 6%.

Versuch 2. Traubenzucker.—Der Organismus lebte in 1-7% igen Lösungen. Vermehrung und Vergrösserung der Vacuolen sehon bei 2% und Abrundung des Körpers bei 3%. Bei einer 4,5%-Kultur wurde die Multiplication sehr verzögert, bei 6% lebte am Ende des fünften Tages noch eine kleine Anzahl der Thiere; bei 7% waren nur noch vereinzelte Individuen am Leben, welche schliesslich nach 5 Tagen abstarben.

Versuch 3. *Milchzucker*.—1-10% ige Lösungen wurden vertragen. Vacuolenvergrösserung von 3% an aufwärts und Körperabrundung über 4%. Bei einer 7% igen Lösung wurde die Vermehrung verzögert, und bei 10% konnten nur einige Indi-

<sup>1)</sup> Figuren in O. Bütschli. loc. cit. 1887-89. Abt, III. Taf. LXII.

viduen 10 Tage lang leben. Auch hier fand mit der Concentrationssteigerung Grössenzunahme des Körpers statt.

Versuch 4. Glycerin.—Maximalconcentration 5%. Vermehrung und Vergrösserung der Vacuolen bei 2%; Körperabrundung bei 3%. Bei 5% lebten nur noch vereinzelte Individuen wenige Tage lang.

Versuch 5. Schwefelsaures Magnesium.—Anpassungsconcentration: 1-5%. Vacuolenvergrösserung und Körperabrundung begannen bei 2%. Bei schwächeren Concentrationen gedieh der Organismus gut, aber über 3% schlecht.

Versuch 6. Salpetersaures Kalium.—Maximalconcentration 2%. Das Thier gedieh bei 0,8% iger Lösung nicht mehr. Zahlzunahme und Vergrösserung der Vacuolen waren wie gewöhnlich.

Versuch 7. Salpetersaures Natrium.—Anpassungsconcentration: 0,2-2%. Ueber 0,8% nahm die Vermehrung stufenweise ab. Gestaltänderung u. s. w. waren ähnlich wie in den vorhergehenden Fällen.

Versuch 8. Chlorkalium.—Anpassungsconcentration: 0,2-1,6%. Die höheren Concentrationen über 0,8% verursachten Multiplicationshemmung. Körperabrundung von 0,6% an. Vacuolenvergrösserung fand auch bei stärkeren Lösungen statt.

Versuch 9. Chlornatrium.—Maximaleoncentration 1,5%. Volumenvergrösserung der Vacuolen wie gewöhnlich.

Versuch 10. Chlorammonium.—Der Organismus konnte sich nur äusserst verdünnten Lösungen anpassen. Schon bei 0,2% trat Vacuolenvergrösserung ein, bei 0,8% Bewegungshemmung und Unregelmässigwerden der Körperumrisse. Bei 1%, der höchsten Concentration, welcher das Thier widerstand, waren einige Individuen nach 2 Tagen noch lebendig.

# (e) Paramaecium caudatum Ehrbg.<sup>1</sup>)

Merkmale des Organismus in der normalen Kultur: Körper verlängert, spindelförmig, biegsam. Wimpern überall an der Oberfläche des Körpers, dicht und gleichmässig. Trichocysten senkrecht zur Oberfläche in der unter der Cuticula unmittelbar befindlichen Rindenschicht gelegen. Mund nahe der Mitte der Bauchseite. Schlund ziemlich lang. Zwei contractile Vacuolen am vorderen und hinteren Ende, mit strahligen zuführenden Kanälen. Nahrungsvocuolen vorhanden. Bewegung lebhaft. Vermehrung langsam.

Versuch. 1. Rohrzucker.—Anpassungsconcentration 1-7%. Von 3% an aufwärts bis 7% Zahl- und Durchmesserzunahme der Vacuolen. Ueber 4% Dickwerden des Körpers; bei 7% blieb das Thier noch viele Tage lang lebendig.

Versuch 2. Traubenzucker.—Anpassungsconcentration: 1-5%. Vacuolenvergrösserung bei ca. 2%, Körperabrundung bei 3%. Sonst wie beim vorhergehenden Versuch.

Versuch 3. Milchzucker.—Maximalconcentration 8%. Vermehrung und Vergrösserung der Vacuolen bei 3% u. s. w. In höheren Concentrationen erreichten die Durchmesser der Vacuolen bedeutend grössere Dimensionen, und der Körper erhielt ein fleischiges Aussehen.

Versuch 4. Glycerin.—Anpassungsconcentration 1-3%. In diesem Medium konnte das Versuchsinfusor nicht lange am Leben bleiben. Vacuolenvergrösserung und Körperabrundung wie bei den vorigen Versuchen.

Versuch 5. Schwefelsaures Magnesium.—Maximalconcentration 2,4%. Obgleich der Körper bei 0,2% verlängert war, so wurde

<sup>1)</sup> Figuren in O. Bütschli. loc. cit. 1887-89. Abt. III. Taf. LXIII.

er doch bei 2,4% viel fleischiger, wobei sich auch die Vacuolen vergrösserten.

Versuch 6. Salpetersaures Kalium.—Anpassungsconcentration 0,2-1%. Die durch dieses Medium hervorgebrachten Gestalt- änderungen waren fast dieselben wie die von Colpidium colpoda bei demselben Medium.

Versuch 7. Salpetersaures Natrium.—Maximalconcentration 1,2%. Dickenzunahme des Körpers von ca. 0,7% an. In 1,2% iger Lösung lebten nur noch vereinzelte Individuen mit schwacher Bewegung.

Versuch 8. *Chlorkalium*.—Anpassungsconcentration 0,2-1%. In einer 1%-Kultur starb das Thier nach 3 Tagen gänzlich ab.

Versuch 9. Chlornatrium.—Anpassungsconcentration 0,2-1%. Hier fand mit Concentrationssteigerung auch Abrundung des Körpers statt.

Versuch 10. Chlorammonium.—Maximalconcentration 0,5%. Das Infusor accommodirte sich an dieses Medium am schwersten; in keiner Kultur blieb es lange am Leben.

## Allgemeines und Schlussbemerkungen.

Aus den oben angeführten Versuchen ergiebt sich, dass mit der Steigerung der Concentration unabhängig von der chemischen Beschaffenheit die Cuticularoberfläche der Infusorienkörper einschrumpft, wenn die Organismen plötzlich in das Medium gebracht werden, weil durch concentrirtere Medien das Wasser aus dem Thierkörper herausgezogen wird. Zugleich wird ihre Bewegung, die bisher lebhaft gewesen war, immer langsamer, und nach einem kurzdauernden Zittern an einem Platze kommen die Thiere endlich zum Stillstande. Wenn aber die Concentration des Mediums nicht zu stark ist, so können es die Infusorien ohne grossen Schaden ertragen, und die einmal gebildeten longitudinalen Falten der Cuticularoberfläche verschwinden nach einiger Zeit wieder. Sogar bei concentrirteren Lösungen findet man nicht selten einige Individuen, welche mit der contrahirten, unebenen Körperoberfläche noch einige Tage lang fortleben können.

Je höher die Concentrationen der Medien sind, desto schwerer wird selbstverständlich die Anpassung, und wenn sich schliesslich die Maximumgrenze nähert, so stirbt der grössere Theil der Individuen ab. Im Falle gelungener Anpassung an ein gewisses Medium sieht man stets Volumen- sowie Zahlzunahme der Chromatophoren, Amylumkörner und Nahrungsvacuolen. Gleichzeitig nimmt der Körper selbst an Dicke und Breite zu, dagegen an Länge etwas ab, so dass er ein einigermassen abgerundetes Aussehen erhält. Zugleich ist ausserdem Grössenzunahme des ganzen Körpers wahrnehmbar, wie ich dies ausschliesslich mit Zuckerarten bei Colpidium colpoda, Mallomonas Plosslii und Chilomonas paramaecium nachgewiesen habe.

Als eine allgemeine Regel gilt auch, dass die Vermehrungsfähigkeit bei höherer Concentration stark beeinträchtigt wird. Unsere Versuche mit den Reinkulturen von Euglena viridis, Chilomonas paramaecium und Colpidium colpoda bieten hierfür unzweideutige Beweise dar. Zum Vergleich führe ich einige der bei Schimmelpilzkulturen gewonnenen Erfahrungen an. Eschenhagen<sup>1</sup>) constatirte, dass das Wachsthum einiger Schimmelarten

<sup>1)</sup> F. Eschenhagen. loc. cit. p. 55.

sich durch stärkere Concentrationen des Substrates stark verzögerte; Klebs¹) beobachtete, dass das Auftreten der Konidienträger und die Perithecienbildung von Eurotium repens, die Keimung und die Sporangienbildung von Mucor racemosus durch die Steigerung der Concentration des Mediums retardirt wurden. Gelegentlich fand ich²) auch, dass Aspergillus niger, der in Magnesiumsulfat-Nährlösungen von verschiedener Concentration gezüchtet wurde, nach 4 Tagen verschiedene Grade der Entwickelung zeigte. Der Pilz wuchs in einer 5%-Kultur am besten, minder gut bei 10%, während bei 20% und 30% nur noch eine sehr schwache Entwickelung zu beobachten war. Die weisse Anlage der Konidienfrüchte trat bei 5% und 10% nach 4 Tagen, bei 20% nach 5 Tagen und bei 30% erst nach 6 Tagen ein.

Unter den zehn von mir angewendeten Stoffen—vier organischen und sechs unorganischen Verbindungen—passten unsere Infusorien sich den Zuckerarten am besten an, und wieder unter den Zuckerarten erwies sich der Milchzucker als das beste Anpassungsmedium. Ihm folgt der Rohrzucker in seiner Concentrationshöhe, während der Traubenzucker schon in weit verdünnteren Lösungen auf die Organismen schädlich einwirkt. Glycerin steht als Anpassungsmedium den Zuckerarten sehr nach. Unter den unorganischen Verbindungen, deren Einwirkung stets viel stärker ist als die der organischen Substanzen, ist schwefelsaures Magnesium zur Vermehrung der Infusorien am geeigneststen, während Chlorammonium für ihr Gedeihen das unpassendste

<sup>1)</sup> G. Klebs. Die Bedingungen der Fortpflanzung bei einigen Algen und Pilzen. Jena 1896. pp. 446-535.

<sup>&</sup>lt;sup>2</sup>) A. Yasuda. Ueber den Einfluss verschiedener unorganischer Salze auf die Fortpflanzungsorgane von Aspergillus niger. The Botanical Magazine. Tokyo 1898. Vol. XII, No. 141. p. 370.

Medium ist, und unter den übrigen Chlorkalium eine mittlere Stellung einnimmt.

Die verschiedenen Infusorien zeigten in Bezug auf ihre Anpassungsfähigkeit grosse Unterschiede, und zwar wohnte unter unseren Infusorien Euglena viridis die grösste Widerstandsfähigkeit inne, während Paramaccium caudatum die kleinste Resistenz besass. Folgende Tabelle zeigt die Grenze der Concentrationen, bis zu welcher die Infusorien am Leben blieben:

<i>J.</i>	Stoffe	Milchzucker	Rohrzucker	Traubenzucker	Glycerin	Schwefelsaures Magnesium	Salpetersaures Kalimn	Salpetersaures Natrium	Chlorkalium	Chlornatrium	Chlorammo- nium
For	$^{\mathrm{C}_{12}\mathrm{H}_{22}^{-}\mathrm{O}_{11}}_{+\mathrm{H}_2\mathrm{O}}$	$C_{12}H_{\underline{22}}O_{11}$	$\mathrm{C}_{6}\mathrm{H}_{22}\mathrm{O}_{6}$	$C_3 \Pi_s$	${ m MgSO}_4$	$\overline{\mathrm{KNO}_3}$	$NaNO_s$	KCl	NaCl	$ m NH_4Cl$	
mit 0,1	rationen der Acq. KNO <sub>3</sub> en Lösungen')	5,40	5,13	2,70	1,38	1,80	1,01	0,85	0,75	0,59	0,54
elchen önnen	Englena viridis	17	15	11	6	6	2,4	2	2,8	1,8	1,4
nen, we	Chilomonas paramae- cium	8	7	6	4	3	2	1,2	2	1	0,6
entratio rien anj	Mallomonas Plosslii	9	7	6	4	3,4	1,5	2,6	1,4	1,5	0,8
Maximalconcentrationen, welchen sich die Infusorien anpassen können	Colpidium colpoda	10	8	7	5	5	2	2	1,6	1,5	1
Maxi sich die	Paramac- cium caudatum	8	7	5	3	2,4	1	1,2	1	1	0,5

<sup>&</sup>lt;sup>1</sup>) Hugo de Vries. Eine Methode zur Analyse der Turgorkraft. Jahrb. f. wiss. Bot. 1884. Bd. XIV. pp. 536—537.

Wie aus der vorstehenden Tabelle ersichtlich ist, muss die Wirkung der angewandten Substanzen auf die Infusorien nicht allein dem Grade ihrer Concentration zugeschrieben werden, dagegen zeigt sich ein annäherndes Verhältniss zu den isotonischen Concentrationen jedes Stoffes. Ich sage ausdrücklich "annähernd," weil die wahre Beziehung zwischen beiden durch unsere Versuche noch nicht sicher gestellt ist. Folgende Tabelle dient zum Vergleiche der isotonischen Concentrationen mit den entsprechenden gefundenen Werthen der maximalen Anpassungsconcentrationen<sup>1</sup>):

<sup>1)</sup> Die bisherigen Untersuchungen ergaben in Bezug auf den Zusammenhang zwischen isotonischen Concentrationen und Reaktionsgrösse durchaus negative Resultate. Man vergleiche hierüber B. Stange. *loc cit.* Nr. 22. p. 364, und C. B. Davenport and H. V. Neal. *loc. cit.* p. 579.

		Stoffe,	Rohrzucker	Milchzucker	Tranbenzucker	Glycerin	Schwefelsaures Magnesium	Salpetersaures Kalium	Salpetersaures Natrium	Chlorkalium	Chlornatrium	Chlorammonium
	Euglena riridis	Concentrationen der Stoffe, die mit 15% Rohrzucker isoto- uisch sind	15	15,8	7,9	4	5,3	3	2,5	2,2	1,7	1,6
	Eug	Gefundene Werthe der maximilen Anpassungsconcen- tration	15	17	11	6	6	2,4	2	2,8	1,8	1,4
	Chilomonas vamaceium	Concentrationen der Stoffe, die mit 7% Rohrzucker isoto- uisch sind	7	7,4	3,7	1,9	2,5	1,4	1,2	1	0,8	0,7
	Chilomonas paramaceium	Gefundene Wertle der maximalen Anpassungsconeen- tration	7	8	6	4	3	2	1,2	2	1	0,6
111	nonas slii	Concentration der Stoffe, die mit 7% Rohrzucker isoto- nisch sind	7	7,4	3,7	1,9	2,5	1,4	1,2	1	0,8	0,7
111	Mallomonas Plosslii	Gefundene Werthe der maximalen Anpassungsconcen- tration	7	9	6	4	3,4	1,5	2,6	1,4	1,5	0,8
IV	liam da	Concentrationen der Stoffe, die mit 8% Rohrzucker isoto- nisch sind	8	8,4	4,2	2,2	2,8	1,6	1,4	1,2	0,9	0,8
IV	Colpidium colpoda	Gefundene Werthe der maximalen Anpassungsconcen- tration	8	10	7	5	5	2	2	1,6	1,5	1
V	tecium	Concentrationen der Stoffe, die mit 7% Rohrzucker isoto- nisch sind	7	7,4	3,7	1,9	2,5	1,4	1,2	1	0,8	0,7
"	Paramaecium caudatum	Gefundene Werthe der maximalen Aupassungsconcen- tration	7	8	5	3	2,4	1	1,2	1	1	0,5

Dieselbe Tabelle zeigt auch zugleich, dass die Anpassungsgrenzen unserer Infusorien an verschiedene Concentrationen im Allgemeinen weit niedriger sind als diejenige der niederen Algen und Schimmelpilze. So kann Zygnema nach Klebs<sup>1</sup>) 50% Rohr-

 $<sup>^{\</sup>rm t})$  G. Klebs. Beiträge zur Physiologie der Pflauzenzelle. Berichte der deutsch bot. Gesellsch. 1887. Bd. V. p. 187.

zucker und 20% Glycerin vertragen. Dass dieselbe Alge sich auch einer 6% igen Chlornatriumlösung anpassen kann, ist von Richter<sup>1</sup>) erwiesen worden. Was die Schimmelpilze anbelangt, so zeigen sie ebenfalls eine weitaus grössere Widerstandsfähigkeit gegen starke Concentrationen. Ich gebe hier zum Vergleiche die von Eschenhagen<sup>2</sup>) erhaltenen Ergebnisse wieder.

	Tranbenzucker.	(ilycerin.	Salpetersaures Natrium.	Chlornatrium.
$Aspergillus\ niger$	53%	43%	21%	17%
Penicillium glancus	n = 55	43	21	18 ,,
Botrytis cinerea	51.,	37.,	16.,	12 ,,

Unsere Infusorien zeigen gegen dieselben Stoffe folgendes Verhalten:

Tra	mbenzucker.	Glycerin.	Salpetersaures Natrium.	Chlornatrium.
Euglena viridis	11%	6 %	2 %	1.8%
$Colpidium\ colpoda$	7 .,	ž .,	<u>·)</u> .,	1,5 .,
Mallomonas Plosslii	ti ,.	4	2.6 .,	$1.5 \dots$
Chilomonas paramaeca	ium 6 .,	4 .,	1.2	1 .,
Paramaecium caudatu	ını 5 ,,	3 ,,	1,2.,	1 ,,

Daraus geht hervor, dass die Resistenzkraft unserer Infusorien gegen die angewendeten Stoffe hinter derjenigen der Schimmelpilze weit zurücksteht.

<sup>1)</sup> A. Richter, loc. cit. p. 24.

<sup>2)</sup> F. Eschenhagen, loc. cit. p. 55.

### Zusammenfassung.

- (1) Isotonische Lösungen der in Rede stehenden Substanzen üben auf die von mir geprüften Infusorien nur eine "annähernd" gleichartige Wirkung aus.
- (2) Die Grenzen der Concentration, welcher sich diese Infusorien unter gewöhnlichen Verhältnissen anpassen können, liegen im Allgemeinen weit niedriger als die der niederen Algen und Schimmelpilze; selbst das widerstandsfähigste darunter, Euglena viridis, vermag nur verhältnissmässig schwache Concentrationen zu ertragen.
- (3) Wenn die Organismen plötzlich in Lösungen höherer Concentrationen gebracht werden, so treten erst an der Cuticularoberfläche ihrer Körper longitudinale Falten auf, aber während ihre Anpassung an das neue Medium stattfindet, dehnen sich die Falten allmählich aus, bis sie zuletzt gänzlich verschwinden.
- (4) Die höhere Concentration des Mediums verlangsamt die Vermehrung der Infusorien.
- (5) Durch Steigerung der Concentration des Mediums wird die Bewegung der Organismen vielfach retardirt.
- (6) Bei Zuckerlösungen stärkerer Concentration vergrössern sieh die Körper der Infusorien bis zu einem gewissen Grade.
- (7) Die Vacuolen, Chromatophoren oder Amylumkörner nehmen in dem Masse an Grösse zu, als die Mediumsconcentration steigt.
- (8) Je mehr die Concentration des Mediums zunimmt, desto mehr runden sich die Körper der Organismen ab, und die Körperumrisse werden uneben.
  - (9) Wenn das Maximum für die Accommodation ein nie-

driges ist, so finden die Veränderungen der Körper der Infusorien schon bei niederen Concentrationen des Mediums statt.

(10) Wenn sich die Concentration des Mediums dem Maximumpunkt nähert, so verschmelzen die in den Körpern der Organismen befindlichen Chromatophoren oder Amylumkörner mehr oder weniger mit einander.

Tokyo, 30. November 1898.

#### Inhaltsübersicht.

EinleitungSeite	101
Litteratur,	103
Methodisches,	107
Beschreibung der Versuche,	112
Allgemeines und Schlussbemerkungen "	127
Zusammenfassung,	134



# Erklärung der Figuren.

Sämmtliche Figuren wurden nach den lebendigen Thieren in den unreinen Kulturen unmittelbar skizzirt, weil ihre Gestalten bei den getödteten Individuen sich mehr oder weniger veränderten.

## TAFEL X.

I	Fig.	1-46.	Chilomo	nas pe	uramaecium	Ehr	rbg.	Ver	gr. 420.
Fig.	1.	Individ	uen aus	einer	Nährlösung	mit	1	% ]	Iilchzucker.
,,	·2.	,,	,,	,,	••	,,	2	••	٠,
,,	3,	,,	,,	••	,,	٠,	3	,,	,,
.,	4.		••	**	•••	••	4	• •	**
٠,	٠,	••	••	••	••	••	.)	٠,	
• •	6.	••	••	••	••	••	6	**	**
••	7.	,•	٠,	,	••	٠,	7		٠,
••	8.	•••		••	••	٠,	8	••	,,
••	9.		••	••	••	••	I	% 1	Rohrzucker.
••	10.		**	••	**	••	• ]	**	
,,	11.		••			٠,	3	••	••
••	12.		**	••	٠,	••	4	••	,,
• •	13,	• •	٠,	• •	**	••	5	••	**
,.	14.	. ,,	,,	,,	1;	••	6	,,	**
٠,	15.	,,	••	**			7	••	**
••	16.	,,	,,	"	,.	٠,	l	%	l'raubenzucker.
••	17.	,,	••	••	••	••	2	••	21
• •	18.	• •	٠,	**	٠,	••	3	,,	**
••	19.	**	٠,	••	•7	71	4	••	• •
7.7	20.	**	• •	٠,	1;	"	5	"	22
,•	21.	,•	••	**	"	••	6		,,
٠,	22.	••	,•	11	••,	٠,		% (i	lycerin.
••	23.	**	*;	• •	**	,,	2	,,	٠,
٠,	24.	,•	**	••	٠,	"	3	,,	,,

Fig.	25,	Individuen	aus	einer	Nährlösung	mit	4 %	Glycerin.
,,	26,	,,	,,	,,	,,	,,	0,5%	Schwefelsaures
								Magnesium.
,,	27.	**	٠,	,,	٠,	,,	1	,,
,,	28.	,,	٠,	,,	,,	,,	1,5 ,,	"
,,	29.	٠,	,,	,•	,.	,,	2 ,,	,,
٠,	30.	••	,,	,,	,,	,,	2,5 ,,	٠,
,•	31.	,,	,,	,.	**	,,	3 .,	,,
٠.	32.	,,	,,	••	,.	••	0,5%	Salpetersaures
								Kalium.
,•	33.	"	٠,	٠,	"	,,	1 ,,	,,
",	34.	,,	"	,,	,,	,,	1,5 ,,	;,
"	35.	**	••	••		• •	2 ,,	"
٠,	36.	,•	٠,	"	"	,,	0.6%	Salpetersaures
								Natrium.
••	37.	٠,	,.			٠,	1,2 ,,	"
٠,	38.	**	••		**		0,5%	Chlorkalium.
,.	39.	,,	••	••	••	,,	1 ,,	.,
••	40.	,,	,.	,.	••		1,5 .,	,,
٠,	41.	.,		٠,			2 ,,	
• •	<b>4</b> 2.	,,	.,		••	• •	0,5%	Chlornatrium.
٠,	<b>4</b> 3.	,,	•,	,.	,,	,,	1 ,,	,,
••	44.		,,	••	**	••	0,2%	Chlorammonium.
.,	45.	••	٠,	••	,•	,,	0,4 ,,	,,
,,	46.	• •	,,	.,	*,	,,	0,6,.	22

### TAFEL XI.

Fig. 1-11. Euglena viridis Ehrbg. Vergr. 420.

Fig. 1. Individuum aus einer Nährlösung mit 1 % Traubenzucker, ,, 2. ,, ,, ,, ,, ,, ,, ,, ,, ,, ,,

,, 3, ,, ,, ,, ,, 3, ,, ,, ,, 4. ,, ,, ,, ,, ,, 4, ,, ,,

Fig.	5.	Individuum	aus	einer	Nährlösung	mit	5	%	Traubenzucker.
,,	6.	"	,,	,,	,,	,.	6	,,	,,
,,	7.	"	,,	,,	,,	,,	7	,,	"
,,	8.	;;	,,	,,	,,	;;	8	,,	,,
"	9.	,,	"	,,	,,	,.	9	;•	22
,,	10.	,,	,,	,,	;;	,,	10	,,	,,
,,	11.	,,	,,	,,	,,	,,	11	,,	,,
	121.	g. 12-41. (	Y . 1	1. 1.000	uolueda Eh	rlw	V		r 190
TN:		g. 12-41. C Individuum							Milchzucker.
Fig.		matyaduum	aus	emer	Nampsung				
"	13.	"	;;	;;	,,	"	2 3	,,	"
٠,	14.	,,	"	"	,,	٠,		"	,,
"	15.		"	;;	**	**	4	,,	٠,
;;	16.	,•	,,	,,	,.	,•	5	22	"
,,	17.	٠,	,,	,,	,,	"	6	"	,,
"	18.	,,	,,	,,	27	,.	7	,.	,,
"	19.	,,	,,	,,	"	••	8	,,	"
,,	20.	,,	,,	"	,,	"	9	,,	,,
,,	21.	;;	٠,	22	,,	,,	10	,,	,,
,,	22.	,,	,,	,,	,,	,,	1	%	Rohrzucker.
,,	23.	,,	,,	,,	٠,	٠,	2	;;	,,
,,	24.	,,	٠,	**	••	.,	3	,,	٠,
,,	25.	,,	,,	,,	,,	,,	4	,,	,,
٠,	26.	27	,,	,,	,,	,,	5	,,	22
,,	27.	,,	,,	,,	,,	22	6	,,	2.2
,,	28.	,,	,,	"	,,	,,	7	٠,	27
,,	29.	,,	,,	,,	,,	,,	8	,,	"
,,	30.	,,	,,	,,	"	,,	1	%	Traubenzucker.
,,	31.	,,	,,	,,	,,	,,	2	,,	,,
٠,	32.	••	,,	,,	,,	,,	3	,,	,,
"	33.	,,	,,	,,	,,	,,	4	"	,,
,,	34.	,,,	,,	,,	,,	,,	5	,,	27
	35						6		

Fig. 36.	Individuum	aus	einer	Nährlösung	$_{ m mit}$	7	% Traubenzucker.
----------	------------	-----	-------	------------	-------------	---	------------------

,,	37.	,,	;;	,,	,,	,,	1	% Glycerin.	

## TAFEL XII.

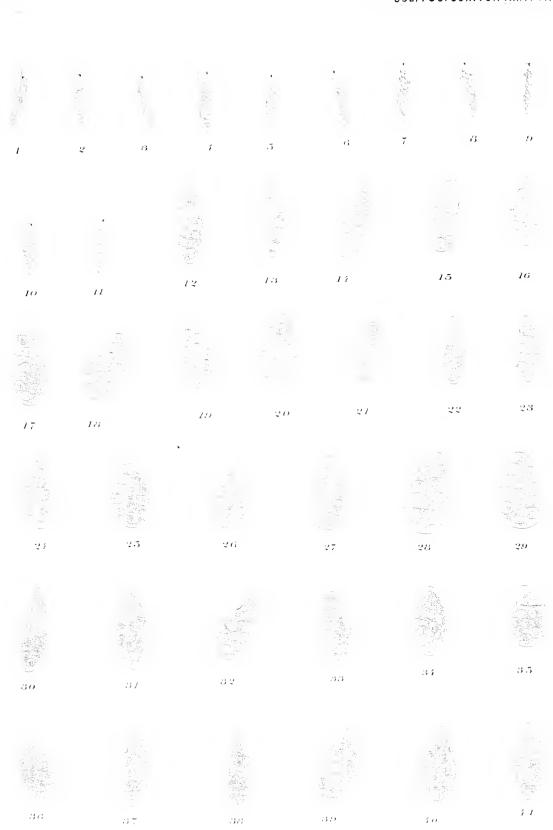
Fig. 1-21. Mallomonas Plosslii Perty, Vergr. 420.

	,							C.	
Fig.	1.	${\bf Individuen}$	aus	einer	Nährlösung	mit	1	00	Milchzucker.
,,	2.	,,	,,	,,	,,	,,	2	,,	"
,,	3.	,,	,,	٠,	,,	,,	3	,,	<b>,</b> ,
,,	4.	,,	.,	,,	"	,,	4	,,	,,
,,	5.	,,	,,	,,	,,	,,	5	,,	"
,,	6.	,,	,,	"	"	,,	6	,,	"
,,	7.	,,	,,	11	<b>31</b>	٠,	7	,,	,,
"	8.	"	,,	,,	,,	,,	8	,,	"
,,	9.	,,	,,	٠,	,,	,,	9	,,	,,
,,	10.	,,	,,	,,	,,	,,	1	%	Rohrzucker,
,,	11.	,,	,,	٠,	,,	٠,	2	,,	,,
,,	12.	,,	,,	,,	,,	,,	3	,,	"
"	13.	"	"	;;	,,	,,	4	,,	,,
,,	14.	"	,,	"	,,	,,	5	,,	,,
,,	15.	"	,,	,,	,,	,,	6	,,	,,
,,	16.	"	,,	,,	,,	,.	7	,,	,,
,,	17.	,,	,,	,,	,,	,,	1	%	Traubenzucker.
,,	18.	,,	27	"	,,	,,	2	,,	,,
"	19.	,,	,,	,,	"	,,	3	,,	,,
,,	20.	"	,,	;;	"	,,	4	,,	"

21.

"

	Fig.	22-28. Par	am	aeciur	n caudatum	Eh	rbg		Verg. 240.
Fig	g. 22.	Individuum	aus	einer	Nährlösung	$_{ m mit}$	1	%	Rohrzucker.
,,	23.	,,	,,	,,	,,	,,	2	,,	,,
,,	24.	,,	,,	,,	,,	٠,	3	,,	"
,,	25.	,,	,.	••	,,	,,	4	,,	,,
,,	26.	<b>,.</b>	,,	,.	,•		5	,,	,,
,,	27.	,,	,,	,,	,,	,,	6	,,	,,
,,	28.	••					7		



24 25 26 27



# Ueber die Wachsthumsbeschleunigung einiger Algen und Pilze durch chemische Reize.

VON

N. Ōno, Rigakushi.

Hierzu Tafel XIII.

## I. Einleitung und Litteratur.

In seiner Arbeit: Études chimiques sur la végétation¹), hat Raulin schon im Jahre 1869 darauf aufmerksam gemacht, dass Zink- und Siliciumsalze in geeigneter Dosis das Wachsthum von Aspergillus niger befördern. Auf dieser an sich richtigen Beobachtung fussend war der genannte Autor mit Unrecht der Ansicht geneigt, dass diese Substanzen zur normalen Entwickelung unseres Pilzes nothwendig seien, indem er diese unter "les éléments chimiques essentiels" rechnet und eine Nährlösung von recht complicirter Zusammensetzung für Pilze vorschreibt.

Ueber die Mineralstoffbedürfnisse der Pilze wurden seither von einigen Forschern Untersuchungen gemacht, von denen die Arbeit Naegeli's<sup>2</sup>) in der ersten Linie zu nennen ist. In

Ann. d. Sc. nat. Bot., Ser. V, T. XI., 1869, S. 91.

<sup>2)</sup> v. Naegeli, Der Ernährungschemismus der niederen Pilze. Sitzungsberichte d. Kgl. Bayr. Akad. d. Wiss. Math.-phys. Cl. 1880.

neuerer Zeit wurde dies Thema von Molisch<sup>1</sup>), sowie auch von Benecke<sup>2</sup>) wiederaufgenommen. Der Mühe dieser Autoren verdanken wir unsere heutigen Kenntnisse in dieser Richtung. Nach den übereinstimmenden Angaben der genannten Autoren stellt weder Zink noch Silicium einen eigentlichen Nährstoff dar und kann wohl von Kulturflüssigkeit ausgeschlossen werden.

Dass die wachsthumsbeschleunigende Wirkung gewisser Metallradikale auf einer chemischen Reizung beruht, wurde von Pfeffer") in "seiner im Jahre 1895 erschienenen Arbeit zum ersten Male klar gestellt und später in allgemeinen Zügen in der 2<sup>ten</sup> Auflage seiner Pflanzenphysiologie erörtert. Er bemerkt in dem letztgenannten Werke, dass anscheinend geringfügige Umstände in der That nicht selten einen erheblichen Einfluss auf Gedeihen und Wachsen haben, und sagt: "Vermuthlich handelt es sich in dieser beschleunigenden Reizwirkung um eine der mannigfachen Reaktionen, die darauf abzielen, durch intensitivere Thätigkeit einen benachtheiligten Einfluss thunlichst entgegenzuarbeiten oder Schädigungen auszugleichen."1) Im vorhergegangenen Jahre wurde eine Reihe Versuche von Richards5) angestellt, deren Resultate die Ansicht Pfeffer's bestätigen. Er zog zu seinen Untersuchungen verschiedene Schwerenmetallsalze wie Zink-, Kobalt-, Nickel-, Eisen-, und Mangansalze und einige andere giftige Substanzen heran und stellte die Thatsache fest, dass fast

<sup>1)</sup> H. Mollisch, Die mineralische Nahrung der niederen Pilze. Sitzungsberichte d. Wiener Akad., Oct. 1894.

<sup>2)</sup> W. Benecke, Die zur Ernährung der Schimmelpilze nothwendigen Metalle, Pringsh. Jahrb. f. wiss. Pot. Ed. XXVIII, 1895, S. 487.

Pfeffer, Election organischer N\u00e4hrstoffe. Pringsh. Jahrb. f. wiss. Bot. Ed. XXVIII, 1895.

<sup>4)</sup> Pflanzenphysiologie, 2 Aufl. Dd. I. S. 374.

<sup>5)</sup> H. M. Richards, Die Beeinflussung des Wachsthams einiger Pilze durch chemische Reize. Pringsh. Jahrb. f. wiss. Bot. Ed. XXX, 1897, S. 665.

alle geprüften Substanzen mehr oder weniger die Pilzernte zu vermehren vermochten.

Auch underweitige Beispiele für Erhöhung der Lebensthätigkeiten durch geringe Zusätze giftiger Stoffe findet man in der Litteratur. So versuchte Schulz¹) zu zeigen, dass die durch Saccharomyces verursachte alkoholische Gährung bei Gegenwart von geringeren Quantitäten gewöhnlich als Hefegifte sich verhaltender Substanzen wie Sublimat, Jod, Salicylsäure, Brom, Arsenige Säure u. a., auf längere oder kürzere Zeit wesentlich gehoben werden konnte. Diese Thatsache aber war bereits gewissen Gewerben nicht unbekannt gewesen, dass Zuführung von sonst gährungshemmenden Stoffe, wie z. B. Kupfervitriol oder Salicylsäure, unter Umständen die Hefe zu energischer Thätigkeit veranlasse. Derselbe Autor hat früher eine ähnliche Erhebung der Lebensäusserungen bei dem thierischen Organismus beobachtet und folgenden Satz ausgesprochen, dass "Jeder Reiz auf eine einzelne Zelle sowohl wie auch auf aus Zellengruppen bestehenden Organen entweder eine Vermehrung oder eine Verminderung ihrer physiologischen Leistungen bedinge, entsprechend der geringeren oder grösseren Intensität des Reizes"2).

Derartigen Verallgemeinerungen begegnen wir auch bei Hueppe<sup>3</sup>). Hauptsächlich auf die auf bakteriologischem Gebiete constatirten Thatsache sich stützend verkündigt er die Erscheinung als den Ausdruck des allgemein giltigen Gesetzes für die Wirkung von Chemikalien auf Protoplasma. Er bezeichnet dieses als das "Biologische Grundgesetz," welches er in folgendem

<sup>1)</sup> H. Schulz, Ueber Helegifte. Pilüger's Archiv f. Physiologie. Ed. 42., 1888. S. 517.

H. Schulz, Zur Lehre von der Arzneiwirkung. Virchow's Archiv. Bd. 108, 1877.
 427.

<sup>3)</sup> F. Hueppe, Naturwissenschaftliche Einführung in die Bakteriologie. 1896, Wiesbaden. S. 55.

Satze formulirt: "Jeder Körper, der in bestimmter Concentration Protoplasma tötet und vernichtet, in geringeren Mengen die Entwickelungsfähigkeit aufhebt, aber in noch geringeren Mengen, jenseits eines Indifferenzpunktes, umgekehrt als Reiz wirkt und die Lebenseigenschaften erhöht."

Zu erwähnen ist noch, dass auch bei höheren Pflanzen dieselbe oder eine wenigstens sehr nahe verwandte Erscheinung vorkommt. Bekanntlich pflegt man seit einigen Jahren die Weinreben mit Kupferpräparaten, der sogenannten Bordeauxbrühe, zur Bekämpfung der Pilzkrankheit zu bespritzen. Eine derartige Behandlung ruft ausser der indirekten Einwirkung, die Schädigung durch parasitische Pilze herabzusetzen, auch eine Schar auffallender Erscheinungen seitens der bespritzten Pflanze hervor, die mit Rumm¹) vielmehr als direkte Wirkung der angewandten Chemikalien auf den Pflanzenorganismus selbst zu bezeichnen Solche sind die Steigerung der Chlorophyllbildung und sind. daraus resultirende vermehrte Stärkeproduktion, reichlicherer Traubenansatz, Beschleunigung der Reifung u. a. Rumm ist der Ansicht, dass die Steigerung der Chlorophyllbildung einem chemischen Reiz zuzuschreiben sei. Im darauf folgenden Jahre führten Frank und Krüger<sup>2</sup>) einige Bespritzungsversuche an Kartoffeln aus, wobei sie auch eine ähnliche Thatsache constatirten. Ueber das eigentliche Wesen der Wirksamkeit jener Stoffe ist vorläufig nichts weiteres zu sagen.

Wie aus der oben angeführten Skizze ersichtlich ist, bezogen

<sup>1)</sup> Rumm, Ueber die Wirkung der Kapferpräparate bei Bekämpfung der sogenanuten Blattkrankheit der Weinrebe, Ber. d. deutsch. Bot. Ges. Bd. XI. 1893, S. 709.

Rumm, Zur Frage nach der Wirkung der Kupfer-Kalksalze bei Bekämpfung der Peronospora viticola, ebenda S. 445.

<sup>2)</sup> Frank u. Krüger, Ueber den Reiz, welchen die Behandlung mit Kupfer auf die Kartoffel hervorruft. Ber. d. deutsch. Bot. Ges. Bd. XII, 1894.

sich, wenn wir von der zuletzt besprochenen Thatsache absehen, alle bisherigen diesbezüglichen Versuche mit pflanzlichen Organismen en ausschliesslich auf chlorophyllose niedere Organismen der Pilze. Wenn die erwähnte Erscheinung, wie vielerseits behauptet wird, allgemeine Geltung haben würde, so sollte es a priori zu erwarten sein, dass auch chlorophyllhaltige niedere Organismen in gleichem Sinne reagiren. Dies aber benötigt einer experimentellen Bestätigung.

Herbeigezogen wurden zu meiner Untersuchung verschiedene Schwerenmetallsalze, wie Zink-, Nickel-, Kobaltsulfat u. a., welche auf unsere Versuchsalgen gewisse Reizung auszuüben vermochten. Ferner wurden eine Reihe Parallelversuche mit Pilzen angestellt, deren Ergebnisse die oben genannten Richards'sche Untersuchung bestätigt und erweitert.

Die vorliegende Arbeit wurde auf Veranlassung und unter Leitung von Herrn Prof. Dr. Miyoshi im Botanischen Institut der Kaiserlichen Universität zu Tokyo während des Zeitraums von August 1898 bis Juni 1899 ausgeführt.

Es ist mir eine angenehme Pflicht, meinem hochverehrten Lehrer für die vielseitige Anregung meinen verbindlichsten Dank auszusprechen.

Herrn Prof. Dr. Matsumura sage ich an dieser Stelle auch meinen besten Dank für die Belehrung und das Interesse, welches er meiner Arbeit entgegengebracht hat.

#### II. Methodisches.

Bei unseren Versuchen kommen stets die Reinkulturen in Betracht. Als Kulturgefässe wurden Erlenmeyer'sche Kolben von ca 200 cc Inhalt angewendet, und zwar von gleicher Gestalt und Qualität in einer Reihe von Parallelversuchen benutzt. Nachdem die Gefässe zuerst mit Salzsäure gründlich gewaschen worden waren, wurden sie mit Leitungswasser, dann mit destillirtem Wasser wiederholt ausgewaschen, getrocknet und gebraucht.

Wasser.—Zur Zubereitung der Nährlösungen und zum Auflösen der Reizmittel benutzte ich doppeltdestillirtes Wasser. Das auf übliche Weise gewonnene destillirte Wasser war von Glas zu Glas nochmals destillirt worden.

Chemische Präparate.—Die als Nährstoffe sowohl als auch als Reizstoffe dienenden Chemikalien stammten grösstentheils aus Merck's "garantirt reinen" Reagentien.

Nährlösungen.—Für Algen bediente ich mich der bekannten Knop'schen Lösung<sup>1</sup>), die ich nach folgender Vorschrift bereitete:

(A) 
$$MgSO_4 + 7H_2O = 10.25g = (B) Ca(NO_3)_2 = 20.00g$$
  
 $KNO_3 = 5.00$ ,  
 $KH_2PO_4 = 5.00$ ,  
Wasser = 175.00cc = Wasser = 175.00 cc

Beim Gebrauch wurden je 10 cc von A und B mit 880 cc Wasser verdünnt. Diese bezeichne ich als Original-Nährlösung für Algen.

Die Original-Lösungen für Pilzkulturen bestanden aus folgenden drei Serien:

<sup>1)</sup> Aus keinem besonderen Grunde benutzte ich hier Ca-haltige Nährlösung. Die Entbehrlichkeit der genannten Metalle bei Pilzen und niederen Algen ist bekanntlich in neuerer Zeit von Molisch und Benecke erwiesen worden.

(A)

$\mathrm{KH_{2}PO_{4}}$	0.50 g
${ m MgSO}_{\scriptscriptstyle 4}$	$0.25^{\circ}$ ,,
$\mathrm{NH_4NO_3}$	1.00 ,,
Eisen	Spuren
Rohrzueker	5.00 g
Wasser	90.00 CC.

(B)

Wie bei A. Asparagin 0.5g statt NH<sub>4</sub>NO<sub>3</sub> 1.0 g

(C)

Wie bei A. Dextrose anstatt Rohrzucker.

Bei fast allen Kulturreihen wurde A angewendet, während B und C nur ausnahmsweise benutzt wurden.

Kulturaustellung.—Ich goss in 5Kolben je 135 cc Original-Lösung (Knop'sche bezw. Rohrzuckernährlösung). Sodann setzte ich zum ersten Kolben 15 cc destillirtes Wasser hinzu und liess dies als Nährlösung für Controlkulturen dienen, zum 2<sup>ten</sup>, 3<sup>ten</sup>... 5<sup>ten</sup> je 15 cc betreffend verdünnte Lösung von Reizstoffen, deren Wirkungen versucht werden sollten. Bei den Algenkulturen geschah die Verdünnung in absteigender geometrischer Reihe im Verhältniss 1:5, bei den Pilzkulturen aber mit 1:2. Alle Kolben wurden darauf gut geschüttelt, um die Lösungen aufs innigste zu mischen, und dann die in jedem Kolben enthaltene Lösung in drei Kolben gleichmässig vertheilt, so dass wir 3 Serien von je 5 Kolben, deren jede 50 cc Nährflüssigkeit enthielt, vor uns haben.

Die auf diese Weise zubereitete Kulturflüssigkeit enthielt bei der Knop'sche Lösung ca 2.5% wasserfreie Salze und bei der Pilznährlösung etwa 5% Rohrzucker¹). Dann folgte bei Pilzkulturen die Sterilisation in einem Koch'schen Dampftopf, welche ¹/2—1 Stunde dauerte.

Bei Pilzen fand die Impfung in üblicher Weise statt, während ich sie bei Algen in der Weise ausführte, dass ich mittelst Platindraht oder Pipette eine möglichst kleine Algenmenge aus den zuvor in Nährlösung von derselben Concentration oder auf Agar bereiteten Reinkulturen herausnahm und in Versuchsgefässe brachte.

Die Kulturen wurden in Zimmertemperatur (ca 15° C im Mittel) ausgeführt, und in kälteren Jahreszeiten ins Treibhaus (16-21° C) gebracht.

Die Kulturdauer variirte unter Umständen zumeist zwischen 8 und 25 Tagen bei Pilzen und etwa einem Monatlang bei Algen.

Bestimmung des Trockengewichtes.—Für die Beurtheilung des Gedeihens hat fast stets die Ermittelung des Trockengewichtes der gebildeten Algen- bezw. Pilzmassen Aufschluss gegeben. Die Bestimmung wurde folgendermassen ausgeführt. Nach Beendigung der Versuche wurde die Kulturflüssigkeit mit der Erntemasse insgesammt durch vorher einzeln gewogene Filter filtrirt. Dabei befreitete ich den an der Glaswand haftenden Theil mittelst eines mit einem Kautschuk-Hut versehenen Glasstäbehens. Dann spülte ich die Erntemasse mit kaltem destillirtem Wasser, um dadurch etwa noch vorhandene Nährflüssigkeit möglichst zu entfernen, und wenn sie ziemlich lufttrocken geworden war, trocknete ich sie im Paraffinofen bei 100° C und wog sie nach dem Erkalten. Das auf diese Weise ermittelte

<sup>1)</sup> Diese Lösung wurde von Pfeffer und Richards vielfach benutzt.

Trockengewicht der Ernte ist in den angeführten Tabellen als Ernteertrag notirt.

Bei den Algenkulturen geschah es vielfach, zumal bei denjenigen, welche während der külteren Jahreszeit angestellt worden waren, dass die Vermehrung nur sehr langsam vor sich ging, und dass nach monatelangem Stehenbleiben eine nur schwache Entwickelung sich zeigte. In solchen Fällen musste ich mich damit begnügen, durch das Aussehen der Kulturen die Stärke der Entwickelung zu beurtheilen.

Was nun die specielle Ausführungsmethode anbelangt, so wird sie an geeigneten Stellen berücksichtigt.

## III. Vorbemerkungen uber Versuchsobjekte.

Für Pilzkulturen bediente ich mich der gewöhnlichen Schimmelpilze Aspergillus niger und Penicillium glaucum.

Benutzt wurden bei meinen Algenversuchen die folgenden Formen:

Protococcus sp.
Chroococcum sp.
Hormidium nitens.
Stigeoclonium sp.

Da wir zur Zeit über die Lebensbedingungen der Algen überhaupt nur wenig wissen, so war es mir nicht immer gelungen, die im Freien rasch wachsenden Algenarten im Laboratorium unbeschädigt gedeihen zu lassen. Besonders schwierig war die Aufgabe, die grösseren Formen in reinem Zustande längere Zeit in einer bestimmten Nährflüssigkeit zu kultiviren.

Nach einigen darauf bezüglichen Vorversuchen kam ich

schliesslich auf die oben genannten niederen Formen zurück, die ziemlich leicht rein zu erhalten waren, ausserdem sicheres Gedeihen zeigten und ferner leichtere Kontrolle der Impfmasse gestatteten.

Unsere kleineren Algen waren zumeist in den für grössere Algen bezweckten Kulturgefässen spontan aufgetreten, und so wurden diese durch wiederholte Uebertragung rein gezüchtet. Die ersteren Algen liessen sich auch auf festem Nährboden, welcher aus ½ Proc. Agar und 2.5 Promille Nährsalz enthaltender Gallerte bestand, gut vegetiren und solcher Plattenkultur bediente ich mich bei einigen Beimpfungen.

Hormidium nitens, welches sich auf der Oberfläche einer Vaucheria-Kultur als eine charakteristische seidenglänzende Decke bildete, zerfiel nach dem Uebertragen in unsere Nährlösung in einzelne Zellen und vegetirte als solche weiter.

Es bleibt noch zu bemerken übrig, dass die Vermehrung bei niederen Algen (*Protococcus* wurde zunächst untersucht) während der kälteren Jahreszeit so gut wie vollständig herabgesetzt worden ist, wenn auch die Kulturen im Treibhaus bei 16-20° C sich befanden. Die Ursache wolle man nicht in mangelndem Licht suchen, da dieselben Kulturen mit noch mässigerem Lichtgenuss vor einem gegen Norden gerichteten Fenster eine gute Entwickelung bis Anfang November zeigten. Ob es sich hier etwa um eine Vegetationsperiodicität handelt, beabsichtige ich im kommenden Jahre näher zu untersuchen.

## IV. Die Veränderungen in der Wachsthumsweise und die Correlation zwischen Fortpflanzung und Wachsthum.

Wie wird die Wachsthumsweise einer Pflanze beeinflusst werden, wenn ihr Wachsthum bei Gegenwart von Reizstoffen über die Norm hinaus gesteigert wird? Um diese Frage zu beantworten, wurden bei meinen Untersuchungen einige Beobachtungen gemacht, um dabei auftretende Wachsthumsmodificationen zu kennzeichnen.

Bei von mir untersuchten Algen konnte ich keine bemerkenswerthe Veränderung der Wachsthumsweise beobachten. Die Zellengrösse blieb unverändert; so lag z. B. bei *Protococcus* sp. die Zellengrösse jedenfalls zwischen 7-10  $\mu$ . Sie zeigte ferner keinen Unterschied in Amylumeinschlüssen.

Bei Pilzkulturen liess sich aber die Veränderung in der Wachsthumsweise mehr oder weniger schon makroskopisch erkennen. So war bei den meisten versetzten Kulturen die Beschaffenheit des Mycels ungewöhnlich. Während bei Kontrollkulturen die Hyphe in Nährlösungen durchsichtig und zart waren, bildeten diejenigen der versetzten Kulturen ein dickes, weisses, hautartiges Geflecht, welches bei längerem Stehen sich zu einer aufgerollten Masse umgestaltete.

Auch das Turgorverhältnis in den versetzten und den nicht versetzten Kulturen wurde vielfach studirt, um zu ersehen, ob hier etwa ein nennenswerther Unterschied zwischen beiden vorhanden ist. Meine Versuche ergaben in diesem Punkte kein positives Resultat.

Viel auffallender sind die Beziehungen zwischen Fortpflanzung und Wachsthum.

Bekanntlich stellen das Wachsthum und die Fortpflanzung zwei miteinander in engster Wechselbeziehung stehende Lebensthätigkeiten dar. So kommt es nicht selten vor, dass bei einer Pflanze, die in üppigem Wachsthum begriffen ist, ihre Fortpflanzungsfähigkeit zeitweilig suspendirt wird; und wenn hingegen die Fortpflanzung herabgesetzt worden ist, so schreitet das Wachsthum kräftig fort.

Es war mir daher nicht ohne Interesse, diese Verhältnisse in unserem Falle kennen zu lernen. Ich konnte jedoch das Studium nur bei Pilzen ausführen, nicht aber bei Algen, da meine Versuchsalgen hauptsächlich einzellige Formen waren, die nur durch Theilung sich vermehrten.

Bei Pilzen hingegen war die Wechselbeziehung zwischen der Mycelentwickelung und der Sporenbildung deutlich zu erkennen.

In fast allen Fällen übten die Versuchsstoffe auf die Pilze Sporen- bezw. Conidienbildung verzögernden Einfluss aus. sonders ausgeprägt trat dies bei Zusatz von ZnSO<sub>4</sub> und NaFl Ich konnte vielfach constatiren, dass bei normalen, in Zimmertemperatur (ca 15° C im Mittel) gezüchteten Aspergillus-Kulturen das Mycelium schon nach 1-2 Tagen sich ausbreitete und mit angelegten Sporangienträgern versehen war, deren Köpfe nach weiteren 2 Tagen durch Reifung der Sporen ganz geschwärzt worden waren. Bei den Kulturen mit Zusatz von 0.005 Proc. ZnSO<sub>4</sub> dagegen habe ich selbst nach Verlauf einer Woche vergeblich nach reifen Sporangien gesucht. In den letzteren Fällen fand ich nur auf dem ziemlich stark angewachsenen häutigen Mycel etwas Sporangienträgeranlage mit etwas ausgeschwollenen Köpfchen, nebst einer Anzahl von wohl als Luftmycel anzusehenden Gebilden. Erst nach weiterem einwöchigen Stehen wurden sie mit bräunlich-schwarzen Sporen besetzt.

Die Grösse der Sporen, welche gegen verschiedener Einflüsse sehr empfindlich ist, blieb in unseren Fällen unverändert. Die Beschaffenheit des Myceliums aber war nicht unbedeutend beeinflusst.

Wie erwähnt, fand ich in allen von mir untersuchten Stoffen

mehr oder minder die Tendenz, die Sporenbildung zu verzögern oder wenigstens zu verspäten. Dies gilt ohne weiteres auch für *Penicillium* sowohl, wie für *Aspergillus*.

Am ausgeprägtesten und am schönsten aber konnte ich dieses Verhältnis bei *Aspergillus*-Kultur mit Zusatz von NaFl kennzeichnen.

Ich nehme hier aus meinem Protokolle folgendes Beispiel:-

- I. Normal Ganze Oberfläche mit schwarzen Sporen bedeckt.
- II. 0.0025% NaFl Etwa $^{1}/_{3}$ der Decke mit Sporen bedeckt, die übrigen $^{2}/_{3}$ steril.
- III. 0.005 ,, ,, Nur sehr spärliche Sporenbildung, weiss.
- IV. 0.010 ,, ,, Steril, hautbildend, weiss.
- V. 0.021 ,, Ganz steril, weiss.

(Beobachtet nach 10 Tagen seit der Aussaatzeit der Sporen.)

Alle NaFl enthaltenden Kulturen zeigten eine üppige vegetative Entwickelung des Myceliums und bildeten hautartige Decken.

Nach weiteren zehn Tagen waren die Kulturen wesentlich unverändert im Aussehen. Bei III. sporadisches Auftreten der Sporangienträger mit unreifen Sporen, bei IV. Sterilbleiben der Decke mit einigen haarigen Luftmycelen, bei V immer steril.

Die Trockengewichtbestimmung nach der Beendigung der Kultur zeigte folgendes Resultat:

Eine Photographie am Ende dieser Arbeit veranschaulicht das eben gesagte Verhältnis.

Die Unterdrückung der Sporenbildung in diesem Falle kann allem Anschein nach eher dem direkt hemmenden Einfluss des betreffenden Stoffes zugeschrieben werden, als der durch stärkere Mycelentwickelung hevorgerufenen Correlationserscheinung, welche häufig bei günstigstem Nährboden zu Stande kommt. Um diesen Punkt aufzuklären, stellte ich die Versuche derart an, dass ich ein Stück Mycelium, welches für 19 Tage in 0.015% NaFl Lösung gezüchtet worden war und keine eigentliche Fruktifikation, abgesehen von einigen schon besprochenen haarigen Gebilden, aufnimmt, nach Bespülung mit Wasser in Normallösung brachte. Schon nach 2 Tagen kamen die angelegten Träger zur Reife und schwärzten, während ich auf der noch in 0.015% verbleibenden Decke vergeblich nach den reifen Sporangien suchte. Dieses Resultat spricht ohne weiteres für die gesagte Ansicht.

Es fragt sich nun, ob die stärkere Entwickelung des vegetativen Organs der Pilze bei der Zugabe einer kleinen Dosis giftiger Stoffe nicht eher als ein specieller Fall der Correlationserscheinungen zwischen Fortpflanzung und Wachsthum zu betrachten ist, indem der direkt hemmende Einfluss der Substanzen auf die Sporenbildung durch Correlation die vegetative Funktion befördert. Es ist schon bestätigt wurden, dass Sporenbildung der Pilze durch einige Gifte<sup>1</sup>) viel empfindlicher beeinflusst wird als die vegetative Mycelentwickelung. Fasst man dieses Verhältnis ins Auge, so ist es wohl begreiflich, dass bei einer genügenden Verdünnung der angewandten Stoffe jene Concentration erreicht ist, welche an sich für die Mycelentwickelung unschädlich, aber für die Sporenbildung hemmend wirkt, und dass so infolge der Correlation das Wachsthum des vegetativen Theils ungewöhnlich gesteigert wird. Dieser indirekten Wirkung der betreffenden Stoffe sehreibe ich, ausser dem direkten Reizeffekt, die Wachsthumssteigerung zu.

<sup>1)</sup> O. Loew, Ein natürliches System der Giftwirkungen 1893.

## V. Einfluss der Reizstoffe auf die Betriebsstoffwechsel.

Die Pflanze nimmt durch ihre Lebensthätigkeiten die Nährstoffe auf und verwendet einen Theil derselben zum Aufbauen ihres Körpers, hingegen den anderen Theil zum Oxydationsmaterial, um dadurch die nothwendige Betriebsenergie sich zu verschaffen. Von diesem Standpunkte aus betrachtet, lassen die im Pflanzenorganismus sich abspielenden Stoffumsätze sich, wie bekannt, in zwei Kategorien: Bau- und Betriebsstoffwechsel trennen. Um die Grösse jedes von diesen Stoffwechseln zu ermitteln, hat man einigermassen einen Maasstab. So ist bei der Betriebsstoffwechselthätigkeit das Trockengewicht maassgebend, und für den Betriebsstoffwechsel giebt die Ermittelung der Kohlensäureproduktion, die Ausscheidung gewisser Stoffwechselprodukte, einige Aufschlüsse.

Wie aus der in der Einleitung angeführten Skizze zu ersehen, beziehen sich die bisherigen Untersuchungen über die Erhöhung der Lebensthätigkeiten durch chemische Reize zumeist nur auf Baustoffwechsel. Richards untersuchte in seiner Arbeit nur den Ernteertrag, berücksichtigte aber nicht den Betriebsstoffwechsel. Schulz beobachtete stärkere Entwickelung der Kohlensäure bei Hefen. Beim ersten Anblick scheint dies auf nur erhöhtem Betriebsstoffwechsel zu beruhen, doch blieb hier die Frage immer offen, ob man es in diesem Falle mit der Erhöhung der Gährthätigkeit einzelner Individuen zu thun hat, oder ob diese Erscheinung von der durch die Reizwirkung verursachten Vermehrung der Gährungserreger bedingt sei.

Es fehlen bisher meines Wissens einschlägige Versuche über die Beeinflussung des Betriebsstoffwechsels in Gegenwart von giftigen Stoffen. Irgend ein Beitrag in dieser Richtung durfte wohl nicht ohne Interesse sein.

Für solchen Zweck bieten die Algen keine geeigneten Objekte dar, wohl aber die Pilze, welche sich dafür bequem anwenden lassen.

Einige Pilze, insbesondere Aspergillus niger, produciren eine nicht unbeträchtliche Menge Oxalsäure als Stoffwechselprodukt, wie aus der bekannten Arbeit Wehmer's¹) hervorgeht. Dieses Stoffwechselprodukt bot bei meinen Versuchen einen Angriffspunkt, und so werden eine Reihe Bestimmungen über die Säurenmenge angeführt. Ich muss hier bemerken, dass ich die Säure nur auf titrimetrischen Wege bestimmte. Die Methode ist untauglich, wenn Oxalsäure nicht nur als solche, sondern auch als Salz vorkommt. Wehmer zeigt aber in seiner oben besprochenen Arbeit, dass in NH<sub>4</sub>NO<sub>3</sub>-haltiger Zuckernährlösung Oxalsäure stets als freie Säure bei Aspergillus-Kulturen auftritt und da meine Kulturen hauptsächlich derartige waren, so war die Titration zuverlässig. Dies geschah mit Liquor Alkali Decinormalis und Phenolphthalein als Indikator.

Nachdem ich von der durch Titration ermittelten gesammten Säure die ursprüngliche Acidität der Nährlösung subtrahirt hatte, rechnete ich diese als Oxalsäure um, welche in der angeführten Tabelle gegeben ist<sup>2</sup>).

<sup>2)</sup> Hier werde ich einige Versuche, um die titrimetrisch ermittelten Zahlen mit denjenigen, die gravimetrisch bestimmt waren, zu vergleichen, ausgeführt angeben:—

Säurenmenge	in g in 10 ce Nährfl	üssigkeit.
Zusatz von NiSO <sub>*</sub>	Titration	Gravimetrisch
0	0.042	0.048
0.003%	0.052	0.060
0.007 ,,	0.050	0.058
0.014 "	0.066	0.075
0.028 "	0.064	0.060
0	0.042	0.040
0.003 ,,	0.047	0.048
0.007 ,,	0.052	0.058
0.014 ,,	0.060	0.065
0.028 ,,	0.051	0.054

<sup>1)</sup> Wehmer, Entstehung und physiologische Bedeutung der Oxalsäure im Stoffwechsel einiger Pilze, Bot. Ztg. 1890.

Vergleicht man nun die Säurenmenge bei Gegenwart von Reizmitteln mit derjenigen der Kontrollversuche, so findet man in unseren Versuchen nur mit der einzigen Ausnahme von NiSO<sub>4</sub> stets das Minus im ersteren Falle. Dieses Verhältnis ist ersichtlich aus der Colonne "Säure pro 1g Pilzsubstanz" in den Tabellen. Steigt der Zusatz von Reizstoffen, so wird die Menge der Säure um so kleiner.

Man kann jedoch nicht annehmen, dass die aufgefundene Säurenmenge die sämtliche Menge der ausgeschiedenen Säure darstellte, da bekanntlich die Ausscheidung der Säure mit ihrer Zersetzung Hand in Hand gehen sollte. Daraus geht hervor, dass die Erklärung der besprochenen Verhältnisse nicht einzig in ihrer Art sein kann. Es könnten einige Möglichkeiten, welche für diese Thatsache sprechen, angegeben werden.

Erstens, wenngleich die Oxalsäure als normales Stoffwechselprodukt unseres Pilzes auftritt, ist sie doch als ein Produkt unvollkommener Oxydation anzusehen, und wenn die Stoffwechselthätigkeit auf einmal gesteigert wird, so wird als das Produkt vollkommener Oxydation mehr Kohlensäure entstehen, dagegen weniger Oxalsäure.

Zweitens könnte dieselbe Erscheinung auftreten, falls die einmal entstandene Säure durch Wiederverarbeitung seitens der Pilze verschwindet. Dabei könnte sie entweder als Baumaterial wiederaufgenommen werden oder, ohne wieder den Pilzen nutzbar zu werden, zersetzt werden. Doch, wie schon Wehmer¹) betont hatte, stellt die Oxalsäure einen nur sehr armen Nährstoff für Aspergillus dar, so dass es wahrscheinlich ist, dass sie bei zureichendem Vorrath von guten Nährstoffen wie Zucker²) wohl

<sup>1)</sup> Wehmer l.c.

<sup>2)</sup> Bei meinem Versuche betrug der Zuckergehalt nach Beendigung der Versuche wenigstens 1.5 g in je 50 cc der Kulturslüssigkeit, d. h. ca 3%.

intakt geblieben wäre. Ferner wurde von Wehmer¹) constatirt, dass weder Licht noch tote Pilzmasse allein die Zersetzung der Säure hervorzurufen im Stande sind, wohl aber die Lebensthätigkeiten der Pilze. Es bleibt daher nur die Annahme übrig, dass die Säure durch Steigerung des Betriebsstoffwechsels lebhafterer Zersetzung unterworfen worden sei.

Die dritte Möglichkeit ist schliesslich die, dass diejenigen Stoffe (Kohlenhydrat u. s. w.), welche bei normaler Wachsthumsenergie durch Stoffwechsel z. Th. als Oxalsäure auftreten, bei der infolge der Reizwirkung über Norm gesteigerten Wachthumsthätigkeit nicht als jene Form abgesondert werden, sondern sich gerade in den integrierenden Theil des Pilzkörpers umwandelten, kurz, dass sie als Baustoff verwendet werden.

Von einer anderen Seite müssen wir also dieses Problem angreifen, um zu entscheiden, ob die eine oder andere von diesen Möglichkeiten für unseren Fall zutrifft. Die Ermittelung der Kohlensäurcausscheidung, des ökonomischen Coöfficienten<sup>2</sup>) u. a. wird wohl an diesen Punkt anschliessend oder wenigstens rathgebend sein.

Im Folgenden gebe ich die Resultate meiner Bestimmungen ökonomischer Coëfficienten bei den Kulturen mit Zusatz von ZnSO<sub>4</sub>, bei denen die Erntezunahme stets am auffallendsten war.

Die Bestimmung des Coëfficienten fand in folgender Weise statt:

Die Kulturflüssigkeit wurde zunächst durch andauerndes Kochen mit verdünnter Salzsäure vollkommen invertirt. Dann verdünnte ich diese bis zu etwa ½ % Zuckergehalt. Eine Burette wurde mit der betreffenden Lösung gefüllt.

In einem Kolben mischte ich genau 10 cc Fehling'sche

<sup>1)</sup> Wehmer l.c.

<sup>2)</sup> Man vergleiche hierüber H. Kunstmann, Ueber das Verhältniss zwischen Pilzernte und verbrauchter Nahrung. 1895. (Leipziger Dissertation).

Lösung mit etwa 40 cc Wasser und brachte es zum Sieden; darauf fügte ich die oben genannte Lösung hinzu, bis schliesslich durch vollkommene Reduction des Kupfers zu Kupferoxydul die Flüssigkeit farblos geworden war.

Da unsere Fehling'sche Lösung in 1000 cc 34.64g Kupfersulfat, 174g Kaliumnatriumtartrat und 120g Natriumhydroxyd enthielt, so sollte je 10 cc derselben durch 0.05g Zucker reducirt werden.

Nun kann man leicht durch die Lesung der Burette die Zuckermenge in der Kulturflüssigkeit kennen lernen. Die Differenz zwischen der ursprünglich vorhandenen Zuckermenge in der Kulturflüssigkeit und der zurückbleibenden ergiebt selbstverständlich die verbrauchte Zuckermenge.

Kulturen mit Zusatz von ZnSO<sub>4</sub>.

Aspergillus niger.

Kultu	rdauer 14 Tage.	Zimmerten	Zimmertemperatur.			
Gehalt an ZuSO, (Gew. %)	Pilzernte in g	Verbrauchte Zuckermenge	Ökonomischer Coëfficient d.h. <u>Verbrauch</u> Ernte			
	Ι	•				
O	0.262	1.594	6.1			
0.0037	0.860	2.429	2.8			
0.0074	0.875	2.429	2.8			
0.0148	0.785	2.380	3.0			
0.0297	0.773	2.340	3.0			
	I	I.				
O	0.386	1.707	4.4			
0.0037	0.924	2.463	2.7			
0.0074	0.928	2.463	2.7			
0.0148	0.918	2.448	2.7			
0.0297	0.837	2,480	2.8			

0 [	0.392	1.819	4.6
0.0037	0.910	2.462	2.7
0.0074	0.908	2.456	2.7
0.0148	0.844	2,456	2.9
0.0297	0.827	2.446	2.8

Was sich nun aus diesem Resultate beurtheilen lässt, ist, dass der ökonomische Coëfficient in jedem Falle bei weitem grösser ist in Kontrolle d.h. in nicht zugesetzter Kultur als in zugesetzter. Dieses Verhältnis deutet also an, dass die Pilze bei Anwesenheit von Zinksulfat veranlasst wurden, mit einem verhältnismässig kleinen Verbrauch von Zucker eine bedeutend grössere Körpersubstanz aufbauen zu können. So scheint mir, wenigstens für Zinksulfat, von den oben besprochenen drei Möglichkeiten die dritte die wirkliche zu sein.

## IV. Specielle Besprechungen.

#### ZnSO.

Unter den von Richards geprüften Stoffen übt dieses Salz die stärkste Wirkung aus.

Auch bei unseren Versuchen mit Algen wirkte ZnSO<sub>4</sub> nächst FeSO<sub>4</sub> sehr günstig auf das Wachsthum ein. Schon bei Zusatz von einer minimalen Quantität, wie 0.000016%, nahm die Ernte etwas zu, und dies war noch deutlicher bei 0.00006% bis 0.0003%. Stieg die Concentration auf 0.0016%, so litten die Algen nicht unerheblich, ohne jedoch das Wachsthum ganz herabzusetzen (ef. Tabelle, Algen A. I-IV).

Unsere Versuche mit Pilzen stimmen mit denjenigen von

Richards überein. Bei längerem Stehen wurde der Unterschied zwischen den Versuchs- und Kontrollekulturen recht überraschend (cf. Tabelle. Pilze A. I-III).

Sehr sonderbar trat einmal bei einer Versuchsreihe mit Dextrose es hervor, dass kein nennenswerther Unterschied in der Ernte sowohl, als auch in der Säurequantität sich erkennen liess. Den Grund davon kann ich aber nicht erklären (Tabelle. Pilze. A. IV.)

Die gelbliche Färbung von Nährflüssigkeiten, sowie die Bildung der bräunlichen Sporen in den Versuchskulturen, welche schon von Autoren besprochen wurden, waren hier bemerklich.

Die Säurenmenge nach Beendigung der Versuche war in Versuchskulturen viel kleiner als in Kontrollen (Tab. Pilze. A. I-III.).

#### FeSO.

Richards giebt an, dass dieses Salz erst bei ziemlich grossem Gehalte einen schädigenden Einfluss ausübt.

Meine betreffenden Versuche mit Algen zeigten auch, dass dasselbe noch höhere Concentration im Vergleich zu anderen Schwerenmetallsalzen erträgt. So lag bei Hormidium das Optimum etwa bei 0.0005% und sogar bei einer höheren Concentration wie 0.0126% war der Ertrag noch etwas grösser als bei den Kontrollen (cf. Tab. Algen. B. I-II.).

In einer mit Zusatz von FeSO<sub>4</sub> angestellten *Penicillium*-Kultur trat merkwürdigerweise das ziegelroth gefärbte Mycelium zu Tage.

#### NiSO.

Bei Algen ruft der Zusatz von NiSO, einen befördernden Einfluss hervor. Die optimale Dosis lag etwa zwischen 0.00006 und 0.00012%, während 0.0028% eine beschädigende Wirkung ausübte (Tab. Algen C. I. II.).

Säureproduktion bei Pilzkulturen war hier im Gegensatz zu den meisten Fällen grösser mit der Erhöhung der Zusätzeprocente (cf. Tabelle Pilze. C. 1-III.).

#### CoSO.

Bei Algen scheint dies auch einen begünstigenden Einfluss auszuüben, doch lag der optimale Punkt etwas niedriger als bei NiSO<sub>4</sub>; Optimum etwa bei 0.00012% (cf. Tab. Algen D. I. II.).

Säureerzeugung war wie gewöhnlich kleiner und zwar sehr regelmässig in Versuchskulturen.

#### CuSO.

CuSO<sub>1</sub> wurde von Richards nicht untersucht. Im Jahre 1897 constatirte Günther¹), dass Kupfersalze in grösseren Mengen das Wachsthum der Pilze retardirten, in geringeren Mengen dagegen besseres Gedeihen mit sich bringen. Auch bei meinen mit Aspergillus und Penicillium angestellten Versuchen beobachtete ich dieselbe Erscheinung. Hattori²) fand auch in seinen Untersuchungen über die Giftwirkung der Kupfersalze eine ähnliche Thatsache.

Hier werde ich zwei Beispiele angeben; für näheres verweise ich auf die tabellarische Zusammenstellung (Tab. Pilze. E.).

Gehalt an CuSO <sub>4</sub>	0	0.0015%	0.003%	0.006%	0.012%
Ernteertrag in g	0.273	0.307	0.313	0.324	0.345

Aspergillus niger.

<sup>1)</sup> E. Günther, Beitrag zur mineralischen Nahrung der Pilze. Erlangen 1897.

Hattori, Ueber die Einwirkung des Kupfersulfates auf einige Pflanzen. Manuskript.

Gehalt an CuSO <sub>4</sub>	0	0.0015%	0.003%	0.006%	0.012%
Ernteertrag in g	0.213	0.320	0.338	0.359	0.410

## Penicillium glaucum.

Bei Algen konnte ich dagegen keine Wachsthumsbeförderung nachweisen, wie aus der Tabelle ersichtlich ist. Schon bei 0.00001% steht die Ernte etwas zurück (Tab. Algen E.). Ob bei noch weiterer Verdünnung die wachsthumsbegünstigende Concentration erreicht sein könnte, lasse ich vorläufig unbestimmt.

Die Säurenmenge in Pilzkulturen war kleiner in Versuchskulturen (Tab. Pilze. E.).

## HgCl2.

Es ist von gewissem Interesse, dass dieses heftige Gift in genügender Verdünnung auch das Wachsthum der Pilze befördert. Schulz<sup>1</sup>) giebt an, dass Kohlensäureentwickelung der Hefe in Gegenwart einer kleinen Menge des Stoffes gesteigert wird. Der optimale Zusatz dabei ist etwa 1/500 000.

Was Schimmelpilze betrifft, so findet man in der bisherigen Litteratur nur die Rede von dem schädigenden Einfluss des betreffenden Stoffs. Raulin²) betrachtet z. B. dies mit AgNO<sub>3</sub>, Pt<sub>2</sub>Cl<sub>6</sub> zusammen als das giftige Salz für Aspergillus. Er gibt 1/512 000 als die Grenze der Giftwirkung. In seinem Experimente mit 1/819 200 konnte er jedoch keinen wachsthumsbeschleunigenden Einfluss beobachten. Meines Wissens liegt uns zur Zeit kein Versuch vor, welcher die letztgenannte Thatsache in positivem Sinne zeigt.

<sup>1)</sup> H. Schulz, l.e.

<sup>2)</sup> Raulin l.c. p. 134.

Meinem Versuche nach (cf. Tab. Pilze. F.) tritt schon bei Verdünnung von 0.0017% oder 1/60 000 ziemlich gute Entwickelung von Aspergillus ein. Stieg die Concentration auf 1/30 000, so kam die Entwickelung zum Stillstand. Die Grenze für Giftwirkung liegt zwischen 1/60 000 und 1/30 000.

Das Optimum war sowohl bei *Peincillium* als auch bei *Aspergillus* etwas unter 0.0013%).

Hier gebe ich zwei Beispiele (cf. Tab. Pilze. F. I-VI).

## Aspergillus niger.

Gehalt an $\mathrm{HgCl}_2$	0	0.0003%	0.0007%	0.0013%	0.0027%
Ernteertrag in g	0.261	0.355	0.354	0.509	0.451

## Penicillium glaucum.

Gehalt an HgCl <sub>2</sub>	0	0.0003%	0.0007%	0.0013%	0.0027%
Ernteertrag in g	0.183	0.249	0.213	0.311	0.246

Säureproduction ist hier wie bei den meisten Fällen kleiner in Versuchskulturen als bei Kontrollen (Tab. Pilze. F.).

Auf Algen übte dieses Salz keinen beschleunigenden Einfluss aus, sondern wirkte nur giftig ein. Schon bei 0.00005% war der schädigende Effekt deutlich zu erkennen. Doch weitere Verdünnung durfte ich nicht ausführen, da bei solchen hohen Verdünnungen einige Fehlerquellen als maasgebend auftreten (Tab. Algen F.).

#### LiNO ..

Von Richards wurde LiCl zur Untersuchung herausgezogen

und in diesem Salze ein eine beträchtliche Wachsthumssteigerung hervorrufender Stoff gefunden. Bei meinem Versuche benutzte ich LiNO<sub>3</sub> mit gleichem Resultate (Tab. Pilze G.)

Säureproduktion war hier auch kleiner in Versuchskulturen als in Kontrollen.

Algen zeigten auch eine besseres Gedeihen in zugesetzten Kulturen (Tab. Algen H.).

#### NaFl.

Dieser Stoff übte auch eine beschleunigende Einwirkung auf Algen aus. Der optimale Punkt liegt etwa bei 0.00003%, stieg die Concentration zu 0.00016% bis 0.0008%, so nahm die Ernte etwas ab, war noch grösser als bei Kontrollen. Erst bei 0.0042% steht der Ertrag im Vergleich zur Kontrolle etwas zurück (Tab. Algen G.).

Bei Pilzen beförderte dieser Stoff das Wachsthum (cf. Tab. Pilze H.). Seine Wirkung auf die Sporenbildung wurde sehon im vorstehenden Capitel behandelt.

Die Säurenmenge in der Nährflüssigkeit war wie gewöhnlich kleiner in zugesetzten als in Kontrolle-Kulturen.

#### Arsen.

Von Arsenverbindungen ist arsenige Säure giftig, doch vertragen höhere und niedere Pflanzen viel Arsensäure<sup>1</sup>).

Da Arsenigsäureanhydrid nur schwer löslich ist, so bediente ich mich des arsenigsauren Kaliums.

Bei *Penicillium*-Kultur war kein bedeutender Unterschied der Ernte sowohl als auch in der Säureproduction bemerklich.

<sup>1.)</sup> O. Loew, System der Giftwirkungen.

Merkwürdig war hier eine eigentliche Geruchsentwickelnng in zugesetzten Kulturen¹).

Auf Algen scheinen die genannten Salze etwas Wachsthumsbegünstigend zu wirken. (Tab. Alg. I).

# VII. Schlussbemerkungen und Zusammenfassung der Resultate.

Aus dem Vorstehenden geht zunächst hervor, dass die chlorophyllführenden niederen Organismen wie Algen in ihrem Gedeihen günstig beeinflusst werden durch einen geringen Zusatz von einigen Stoffen, welche für sich nicht Nährstoffe sind, ja sogar giftig wirken. In dieser Reaktion verhalten sich die Algen gerade wie die Pilze. Nur ist zu bemerken, dass die optimale Dosis für Algen viel kleiner als bei Pilzen ist, eine Thatsache, welche vielleicht vom oekologischen Standpunkte aus ihren Aufschluss haben wird. Von den geprüften Stoffen konnte ich nur bei Quecksilberchlorid und Kupfersulfat die besprochene Reaktion nicht constatiren, indem ich bei ihnen, soweit meine Versuche reichten, stets Giftwirkung beobachtete. Daraus muss aber nicht geschlossen werden, dass den beiden Stoffen die nämliche Eigenschaft nicht zukommt, da man bei ihnen unter Umständen doch noch jene wachsthumsbegünstigende Einwirkung wohl erwarten kann.

Bei Pilzen konnte ich die früheren Versuche Richards' hauptsächlich bestätigen, dazu prüfte ich mit positiven Resultaten einige bisher noch nicht untersuchte Stoffe.

Die Verzögerung oder Verspätung der Sporenbildung bei unseren Versuchen ist nicht als infolge einer üppigen vegetativen

<sup>1)</sup> Schon von Gasio (Jahresber, über Gährungsorganismen 1893) erörtert.

Entwickelung verursachte Correlationserscheinung, vielmehr als durch Reizstoffe bewirkte Hemmung zu betrachten.

Was nun die Art und Weise der Reizwirkung anbelangt, so bemerke ich folgendes:

Wenn es sich hier zunächst um zeitliche oder andauernde Hyperaesthesia handelt, so muss Bau- und Betriebsstoffwechsel gleichzeitig gesteigert werden.

Wenn aber dagegen durch Zusätze der Reizstoffe die Thätigkeiten seitens des Organismus so gesteigert werden, dass sie mit kleinerem Energieaufwand die Nährstoffe in sich aufnehmen und sich bauen, kurz, ökonomisch arbeiten können, so kann der dynamische Stoffwechsel nicht so erheblich beeinflusst bleiben. Um daher in dieser Hinsicht eine richtige Aussaung zu gewinnen, ist ein Einblick in den Betriebsstoffwechsel von Wichtigkeit. Einige von meinen Versuchen in dieser Richtung zeigten andeutungsweise, dass Betriebsstoffwechsel nicht parallel mit Baustoffwechsel gesteigert werden; doch sind zur Zeit meine diesbezüglichen Versuche leider unzureichend, um in bezug auf diesen Punkt Allgemeines zu sagen.

Zum Schluss seien im Folgenden die wichtigsten Resultate kurz zusammengestellt:—

- 1. Das Gedeihen der niederen Algen wird durch Einführung gewisser giftiger Stoffe in höchst verdünnten Zuständen begünstigt. Hierzu gehören ZnSO<sub>4</sub>, NiSO<sub>4</sub>, FeSO<sub>4</sub>, CoSO<sub>4</sub>, NaFl, LiNO<sub>5</sub>, K<sub>2</sub>AsO<sub>5</sub>.
- 2. Die Erntezunahme bei Algen muss auf die vegetative Vermehrung der Individuenzahl zurückzuführen sein, da keine nennenswerthe Veränderung der Körpergrösse bemerkbar war.
  - 3. Die geeignete Dosis ist bei Algen bedeutend kleiner als

bei Pilzen. Schon der Zusatz von 10<sup>-4</sup> Gr. Mol. Salz wirkte in den allermeisten Fällen schädigend.

- 4. In CuSO<sub>4</sub> und HgCl<sub>2</sub> fand ich, soweit unsere Studien ausreichen, keine beschleunigende Wirkung auf Algen, wohl aber begünstigend bei Pilzen.
- 5. Bei Pilzen tritt durch Zusätze von HgCl<sub>2</sub> (Optimum etwa bei 0.0013% und CuSO<sub>4</sub> (Optimum etwa bei 0.012%) die Wachsthumsbeschleunigung ein.
- 6. Die Säurequantität in Kulturen mit Zusatz von ZnSO<sub>4</sub>, CoSO<sub>4</sub>, HgCl<sub>2</sub>, NaFl, CuSO<sub>4</sub> war stets kleiner als in Kontrolkulturen. Nur verhielt NiSO<sub>4</sub>, soweit meine Versuche ein Urtheil gestatten, sich diametral entgegengesetzt.
- 7. Die geprüften Stoffe (speciell ZnSO<sub>4</sub> und NaFl) neigen dazu, die Sporenbildung der Pilze direkt zu hemmen, wenigstens das Auftreten der Sporen zu verspäten.
- 8. Die oekonomischen Coefficienten in ZnSO<sub>4</sub>-Kultur sind in der Kontrolle, d. h. in der nicht zugesetzten Kultur, bei weitem grösser als in der zugesetzten.

Juni, 1899.

Botanisches Institut Kaiserl. Universität zu Tokyo.



## TABELLARISCHE ZUSAMMENSTELLUNG.

#### BEMERKUNGEN:

Versuche mit Algen—Der Entwickelungsgrad ist entweder mit Erntegewicht oder mit den relativen Werth gebenden Ziffern bezeichnet.

Versuche mit Pilzen—In Colonne "Acidität" ist die Quantität des Decinormal Alkalis in ee gegeben, welche 10 ee der Nährflüssigkeit neutralisirte (Die ursprüngliche Acidität wurde natürlich vorher subtrahirt). Darans ermittelte ich die Säurenmenge in je 50 ee Nährflüssigkeit, berechnete sie als Oxalsäure und in Colonne "als Oxalsäure umgerechnet" angab.

In Kolonne "Säure pro 1<br/>g Pilzsubstanz" ist das Verhältnis  $\frac{\text{Oxalsaure}}{\text{Ernte}}$ gegeben.

Obwohl bei einigen Penicillium-Kulturen die Säurenmenge gegeben sind, durfte ich doch nicht die  $\frac{Oxalsaure}{Ernte}$  ermitteln, da bei Penicillium das Titration unzuverlässig scheint.

## I. Versuche mit Algen.

A. I. Kulturen mit Zusatz von ZuSO<sub>4</sub>.

Ernteertrag in g		$_{ m n.g.}$ Prot	ococcus sp.—ang	estellt 5. Oct.	Zimmer-Temperatur.		
olt in	Gram Mol.	0	$\frac{1}{2} \times 10^{-6}$	$\frac{1}{2} \times 10^{-5}$	$\frac{1}{2} \times 10^{-4}$	Kultur-	
Gehalt	Gew. %	0	0.000014	0.00014	0.0014	dauer	
	I	0.010	0.018	0.018	0.002	23 Tage	
	II	0.016	0.023	0.018	0.009	26 ,,	
	III	0.012	0.019	0.021	0.006	. ?	

A. II. Kulturen mit Zusatz von ZnSO4.

Ernteertrag in g.			Protococcus sp	.—angestellt	Zimmer-Temperatur.		
ılt in	Gram Mol,	0	$\frac{1}{2.5} \times 10^{-5}$	$\frac{1}{5} \times 10^{-5}$	10-5	$\frac{1}{2} \times 10^{-4}$	Kultu-
Gehalt	Gew. %	0	0.000012	0.00006	0.0003	0.0014	dauer
	I	0.035	0.043	0.038	0.042	0.024	44 Tage
II 0.032 III 0.030		0.032	0.040	0.036	0.040	0.023	46 "
		0.040	0.037	0.042	0.020	50 ,,	

A. III. Kulturen mit Zusatz von ZnSO4. Chrococcum—angestellt 24 Dec

Ernteertrag in g.			Chroococcum—angestent 24. Dec. in Treibhaus 16–20° C.				
alt in	Gram Mol.	0	$\frac{1}{25} \times 10^{-5}$	$\frac{1}{5} \times 10^{-5}$	10-5	$\frac{1}{2} \times 10^{-4}$	Kultur-
Gehalt	Gew. %	0	0.000012	0.00006	0.0003	0.0014	dauer
	I 0.00		0.015	0.026	0.022	0.006	71 Tage
0.009		0.021	0.022	0.024	0.010	,,	
0.010		0.024	0.026	0.026	0.009	,,	

### A. IV. Kulturen mit Zusatz von ZnSO<sub>4</sub>.

	Ernteertrag in g.		Protococcus—angestellt 5. Feb. in Treibhaus 16-20° C.				
alt in	Gram Mol.	0	$\left  \frac{1}{25} \times 10^{-5} \right $	$\frac{1}{5} \times 10^{-5}$	$10^{-5}$	$\frac{1}{2} \times 10^{-4}$	Kultur-
Gehalt	Gew. %	0	0.000012	0.00006	0.0003	0.0014	dauer
	I	0.008	0.017	0.019	0.019	0.007	65 Tage
	H	0.009	0.018	0.023	0.016	0.009	,,
	III	0.011	0.015	0.018	0.017	0.005	,,

## B. I. Kulturen mit Zusatz von FeSO<sub>4</sub>. Hormidium niteus—angestellt 7. Oct.

	Ernteertrag in g. Hormidium nilens—anges				It 7. Oct.	Zimmer-Temperatur.	
alt in	Gram Mol.	0	$\left  \frac{1}{25} \times 10^{-4} \right $	$\frac{1}{5} \times 10^{-4}$	$10^{-4}$	$\frac{1}{2} + 10^{-3}$	Kultur-
Gehalt	Gew. %	0	0.0001	0.0005	0.0025	0.0126	dauer
	I	0.038	0.072	0.074	0.080	0.042	65 Tage
	$\Pi$	0.028	0.083	0.079	0.062	0.041	,,
	III	0.031	0.070	0.082	0.074	0.039	,,

## B. H. Kulturen mit Zusatz von FeSO<sub>4</sub>.

	Ernteertrag in g. Hormidium nitens—angestellt 10. Nov. in Treibhaus 16-							
alt in	Gram Mol.	0	$\left  \frac{1}{25} \times 10^{-4} \right $	$\frac{1}{5} \times 10^{-1}$	$10^{-4}$	$\frac{1}{2} \times 10^{-3}$	Kultur-	
Gehalt	Gew. %	0	0.0001	0.0005	0.0025	0.0126	dauer	
	I	0.025	0.050	0.052	0.054	0.041	79 Tage	
	II	0.023	0.067	0.049	0.042	0.032	,,	
	111	0.027	0.064	0.058	0.046	0.032	,,	

C. I. Kulturen mit Zusatz von NiSO<sub>4</sub>.

	Ernteertrag in g.		Chroncoccum	Chroncoccum—angestellt 2. Oct.			Zimmer-Temperatur.	
ult in	Gram Mol.	0	$\frac{1}{2.5} \times 10^{-5}$	$\frac{1}{5} \times 10^{-5}$	10-5	$\frac{1}{2} \times 10^{-4}$	Kultur-	
Gehalt	Gew. %	0	0.000012	0.00006	0.00028	0.0014	dauer	
	I	0.012	0.012	0.025	0.021	0.004	64 Tage	
	II	0.011	0.015	0.024	0.020	0.006	,,	
	III	0.013	0.018	0.020	0.022	0.007	,,	

## C. H. Kulturen mit Zusatz von NiSO<sub>4</sub>.

Hormidium nitens-angestellt 16. Nov. in Treibhaus 16-24° C.

ılt in	Gram Mol.	0	$\frac{1}{25} \times 10^{-5}$	$\frac{1}{5} \times 10^{-5}$	10-5	$\frac{1}{2} \times 10^{-4}$	Kultur-
Gehalt	Gew. %	0	0.000012	0.00006	0.00028	0.0014	dauer
	Ι	4	5	4-5	4	1	70 Tage
	II	4	5	4	-4	1	,,
	III	3-4	5	4-5	4	1	,,

N.B. Die Ziffer zeigt den Entwickelungsgrad.

### D. I. Kulturen mit Zusatz von CoSO.

Hormidium nitens—angestellt 9. Dec. in Treibhaus 16-24° C.

ilt in	Gram Mol.	0	$\frac{1}{25} \times 10^{-5}$	$\frac{1}{5} \times 10^{-5}$	10-5	$\frac{1}{2} \times 10^{-4}$	Kultur-
Gehalt	Gew. %	0	0.000012	0.00006	0.0003	0.0014	dauer
_	I	4.5	5	4.5	-1	2.5	70 Tage
	II	4	ð	4.5	3.5	2	,,
	III	4	5	4.5	4	2	,,,

N.B. Die Ziffer zeigt den Entwickelungsgrad.

### D. II. Kulturen mit Zusatz von CoSO<sub>4</sub>.

Protococcus—angestellt 19. Mai. Ernteertrag in g. Zimmer-Temperatur. Gram  $\frac{1}{2.5} \times 10^{-5} + \frac{1}{5} \times 10^{-5}$  $10^{-5}$  $\frac{1}{2} \times 10^{-4}$ 0 Kultur-Mol. dauer 0.000012 Gew. % 0 0.000060.00030.0014 Ι 0.0120.0240.026 0.020 0.010 32 Tage  $\Pi$ 0.0090.0300.0250.0220.007Ш 0.010 0.0240.020 0.018 0.006

E. I. Kulturen mit Zusatz von CuSO<sub>4</sub>. Stigeoclonium—angestellt 14. Oct.

ium—augestellt 14. Oct. Zimmer-Temperatur.

alt in	Gram Mol.	0	$\left  \frac{1}{25} \times 10^{-5} \right $	$\frac{1}{5} \times 10^{-5}$	10-5	$\frac{1}{2} \times 10^{-4}$	Kultur-
Gehalt	Gew. %	0	0.00001	0.00005	0.00025	0.0012	dauer
	I	5	4	2	1	1	27 Tage
	II	5	4	2-3	1	1	,,
	III	5	4	2	1	1	,,

N.B. Die Ziffer zeigt den Entwickelungsgrad,

### E. II. Kulturen mit Zusatz von ${\rm CuSO_4.}$

Chroococcum—angestellt 13. Dec.

in Treibhaus 16-20° C.

olt in	Gram Mol.	0	$\frac{1}{25} \times 10^{-5}$	$\frac{1}{5} \times 10^{-5}$	10-5	$\frac{1}{2} \times 10^{-4}$	Kultur-
Gehalt	Gew. %	0	0.00001	0.00005	0.00025	0.0012	dauer
	I	5	4	3	2	0	71 Tage
	II	4-5	4-5	3	2	О	,,
	III	5	4	3	2	0	,,

N.B. Die Ziffer zeigt den Entwickelungsgrad.

### F. I. Kulturen mit Zusatz von $\mathrm{HgCl}_{\,2}.$

Protococcus—angestellt 25. Dec.

in Treibhaus 16-20° C.

alt in	Gram Mol.	0	$\frac{1}{25} \times 10^{-5}$	½ × 10-5	10-5	$\frac{1}{2} \times 10^{-4}$	Kultur-
Gehalt	Gew. %	0	0.00001	0.00005	0.00025	0.00124	dauer
	1	5	4	1	0	0	43 Tage
	II	5	3	0	0	O	,,
	III	5	3	0	0	O	,,

N.B. Die Ziffer zeigt den Entwickelungsgrad

#### G. I. Kulturen mit Zusatz von NaFl.

Ernteertrag in g. Probeoccus—angestellt 24. Dec. in Treibhaus 16-20° C.

alt in	Gram Mol.	()	$_{125}^{1} \times 10^{-3}$	$\frac{1}{25} \times 10^{-3}$	$\frac{1}{5} \times 10^{-3}$	10-3	Kultur-
Gehalt	Gew. %	0	0.00003	0.00016	0.0008	0.0042	dauer
	I	0.012	0.018	0.018	0.018	0.011	76 Tage
	II	0.012	0.025	0.018	0.015	0.015	,,
	III	0.010	0.027	0.015	0.015	0.011	,,

H. I. Kulturen mit Zusatz von  $\mathrm{LiNO}_3.$ 

Protococcus—angestellt 16. April. Ernteertrag in g. Zimmer-Temperatur. Gram  $\frac{1}{25} \times 10^{-4}$   $\frac{1}{5} \times 10^{-4}$ 10-1  $\frac{1}{2} \times 10^{-3}$ 0 Kultur-Mol. Gew. % 0 dauer. 0.000030.000140.0007 0.0034 Ι 0.010 0.020 0.017 0.012 0.009 24 Tage H 0.009 0.0200.0200.015 0.010 III0.010 0.018 0.0160.011 0.008 ,,

I. I. Kulturen mit Zusatz von K3AsO4.

Protococcus—angestellt 24. Dec. Ernteertrag in g. In Treibhaus 16-20° C. Gram  $\frac{1}{125} \times 10^{-4}$   $\frac{1}{25} \times 10^{-4}$  $\frac{1}{5} \times 10^{-4}$ 10-4 0 Kultur-Mol. dauer. Gew. % 0 0.000020.00010.00050.0024Ι 0.011 0.0150.0120.0110.008 54 Tage 0.012  $\Pi$ 0.008 0.017 0.0200.007 III0.009 0.0150.018 0.0140.009 ,,

#### II. Versuche mit Pilzen.

A. I. Kulturen mit Zusatz von ZnSO<sub>4</sub>.

Aspergillus niger—angestellt 24. Dec. '98.

Geerntet 18. Jan. Kulturdauer 25 Tage. Temperatur 16–20° C.

Gelialt in		Ernteertrag	1	Säure pro 1g.		
Gram Mol.	Gram Mol. Gew. %		Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.	
0	0	0.216	15.7	0.495	2.245	
$\frac{1}{8} \times 10^{-3}$	0.003	0.863	12.9	0.406	0.470	
$\frac{1}{4} \times 10^{-3}$	0.007	0.938	13.2	0.416	0.443	
$\frac{1}{2} \times 10^{-3}$	0.014	0.944	12.9	0.406	0.430	
$10^{-3}$	0.028	0.951	13.0	0.409	0.430	

N.B. Asparagin als N-Quelle.

A. II. Kulturen mit Zusatz von ZnSO<sub>4</sub>. Aspergillus niger.—angestellt 24. Dec. '98.

Geerntet 20. Jan. '99.

Kulturdauer 27 Tage.

Temperatur 16-20° C.

Geha	Gehalt in  Gram Mol. Gew. %		Si	iure	Säure pro 1g.
Gram Mol.			Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.181	14.8	0.467	2.580
$\frac{1}{8} \times 10^{-3}$	0.003	0.868	11.8	0.372	0.428
$\frac{1}{4} \times 10^{-3}$	0.007	0.870	11.1	0.350	0.420
$\frac{1}{2} \times 10^{-3}$	0.014	0.858	12.1	0.381	0.444
$10^{-3}$	0.028	0.821	14.1	0.444	0.541

N.B. Asparagin als N-Quelle.

# A. III. Kulturen mit Zusatz von ZnSO . Aspergillus niger—angestellt 24. Dec. '98.

Geerntet 20, Jan. '99.

Kulturdauer 27 Tage.

Temperatur 16–20° C.

Gehal	Gehalt in		S	Säure pro 1g.	
Gram Mol.	Gew. %	in g.	'Acidität	als Oxal-äure umgerechnet	Pilzsubstanz.
0	0	0.187	17.8	0.561	3.000
$\frac{1}{8} \times 10^{-3}$	0.003	1.017	12.5	0.394	0.387
$\frac{1}{4} \times 10^{-3}$	0.007	1.336	11.8	0.372	0.278
$\frac{1}{2} \times 10^{-3}$	0.014	1.939	12.5	0.394	0.419
$10^{-3}$	0.028	1.204	11.2	0.353	0.293

N.B. Asparagin als N-Quelle.

## A. IV. Kulturen mit Zusatz von ZnSO<sub>4</sub>. Aspergillus niger—angestellt 20. Febr.

Geerntet 11. Jan.

Kulturdauer 20 Tage.

Temperatur 16-20° C.

Geha	lt in	Ernteertrag	S	Säure pro 1g.	
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgereehnet	Pilzsubstanz.
O	0	0.634	7.2		
$\frac{1}{8} \times 10^{-3}$	0.003	0.641	7.2		
$\frac{1}{4} \times 10^{-3}$	0.007	0.635	7.2		
$\frac{1}{2} \times 10^{-3}$	0.014	0.627	7.2		
$10^{-3}$	0.028	0.585	7.2		

N.B. Dextrose austatt Rohrzucker.

## B. I. Kulturen mit Zusatz von FeSO<sub>4</sub>. Penicillium glaucum—angestellt 11. April.

Geerntet 25. April.

Kulturdauer 14 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	S	Säure pro 1g.	
Gram Mol.	Gram Mol. Gew. %		Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	O	0.191	3.5		
$\frac{1}{4} \times 10^{-3}$	0.007	0.180	3.3		
$\frac{1}{2} \times 10^{-3}$	0.014	0.233	3.9		
$10^{-3}$	0.028	0.201	3,5		
$2 \times 10^{-3}$	0.056	0.181	3.4		

N.B. NH<sub>4</sub>NO<sub>4</sub> N-Quelle. In allen eisenhaltigen Kulturen waren die Pilzmassen schön ziegelroth gefärbt. Schon bei 0.007 ♯ deutliehe rothe Färbung bemerklich.

#### C. I. Kulturen mit Zusatz von NiSO<sub>4</sub>.

Aspergillus niger—angestellt 9. Febr.

Geerntet 3. März.

Kulturdauer 22 Tage.

Temperatur, 16-20° C.

Gehalt in		Ernteertrag	S	Säure	
Gram Mol.	Gew. %	in g.	$\Lambda { m cidit} { m i}{ m d}{ m t}$	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.250	11.0	0.346	1.464
$\frac{1}{8} \times 10^{-3}$	0.003	0.297	11.1	0.350	1.179
$\frac{1}{4} \times 10^{-3}$	0.007	0.315	10.7	0.337	1.069
$\frac{1}{2} \times 10^{-3}$	0.014	0.401	14.6	0.460	1.147
$10^{-3}$	0.028	0.295	14.0	0.441	1.493

N.B. NH 4 NO 3 als N-Quelle.

### C. II. Kulturen mit Zusatz von NiSO.

Aspergillus niger—angestellt 9. Febr.

Geerntet 4. März.

Kulturdauer 23 Tage.

Temperatur 16-20° C.

Geh	Gehalt in		Si	iure	Säure pro 1g.
Gram Mol.	Gew. %	in g.	Aciditat	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.288	11.1	0.350	1.216
$\frac{1}{8} \times 10^{-3}$	0.0035	0.316	10.7	0.337	1.067
$\frac{1}{4} \times 10^{-3}$	0.007	0.307	11.1	0.349	1.130
$\frac{1}{2} \times 10^{-3}$	0.014	0.387	16.9	0.532	1.375
$10^{-3}$	0.028	0.329	16.7	0.526	1.500

N.B. NH, NO3 als N-Quelle.

C. III. Kulturen mit Zusatz von NiSO,.

Aspergillus niger—angestellt 9. Febr.

Geerntet 6. März.

Kulturdauer 35 Tage.

Temperatur 16–20° C.

Gehalt in		Ernteertrag	S	Säure pro 1g.	
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.324	9.8	0.308	0.951
$\frac{1}{8} \times 10^{-3}$	0.003	0.310	10.0	0.315	0.016
$\frac{1}{4} \times 10^{-3}$	0.007	0.329	11.9	0.375	1.140
$\frac{1}{2} \times 10^{-3}$	0.014	0.362	15.2	0.479	1.323
10-3	0.028	0.341	17.3	0.544	1.695

N.B. NH<sub>4</sub>NO<sub>3</sub> als N-Quelle.

#### C. IV. Kulturen mit Zusatz von NiSO<sub>4</sub>. Aspergillus niger—angestellt 22. April.

Geerntet 4. Mai.

Kulturdauer 12 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.262			
$\frac{1}{8} \times 10^{-3}$	0.003	0.390			
$\frac{1}{4} \times 10^{-3}$	0.007	0.404			
$\frac{1}{2} \times 10^{-3}$	0.014	0.364			
10-3	0.028	0.315			

N.B. NH4NO3 als N-Quelle,

#### C. V. Kulturen mit Zusatz von NiSO<sub>4</sub>. Aspergillus niger—angestellt 22. April.

Geerntet 4. Mai.

Kulturdauer 12 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.214			
$\frac{1}{8} \times 10^{-3}$	0.003	0.311	~		
$\frac{1}{4} \times 10^{-3}$	0.007	0.300			
$\frac{1}{2} \times 10^{-3}$	0.014	0.307			
$10^{-3}$	0.028	0.296			

N.B. NII NO3 als N-Quelle.

C. VI. Kulturen mit Zusatz von NiSO<sub>4</sub>. Aspergillus niger—angestellt 22. April.

Geerntet 4. Mai.

Kulturdauer 12 Tage.

Temperatur 16-20° C.

· Gehalt in			Frnteertrag	Säure		Säure pro 1g.	
Gram Mcl.	Gew. 76	-	in g.		Λeidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	į	0.278				
$\frac{1}{8} \times 10^{-3}$	0.003		0.340				
$\frac{1}{4} \times 10^{-3}$	0.007		0.325				
$\frac{1}{2} \times 10^{-3}$	0.014		0.308				
10-3	0.028		0.324				

N.B. NH4NO3 als N.Quelle.

# D. I. Kulturen mit Zusatz von CoSO<sub>4</sub>. Aspergillus niger—angestellt 17. Febr.

Geerntet 16. März.

Kulturdauer 27 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.297	9.0	0.283	0.953
$\frac{1}{16} \times 10^{-3}$	0.0017	0.439	10.3	0.324	0.738
$\frac{1}{8} \times 10^{-3}$	0.0035	0.565	12.3	0.387	0.685
$\frac{1}{4} \times 10^{-3}$	0.007	0.751	11.2	0.353	0.470
$\frac{1}{3} \times 10^{-3}$	0.014	0.872	8.7	0.274	0.314

N.B. NH NO3 als N-Quelle,

D. II. Kulturen mit Zusatz von CoSO, Aspergillus niger—angestellt 17. Febr.

Geerntet 20. März.

Kulturdauer 31 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	S	Säure		
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.	
0	0	0.280	10.0	0.315	1.125	
$\frac{1}{16} \times 10^{-3}$	0.0017	0.423	12.1	0.381	0.900	
$\frac{1}{8} \times 10^{-3}$	0.0035	0.582	15.5	0.491	0.844	
$\frac{1}{4} \times 10^{-3}$	0.007	0.745	16.7	0.548	0.735	
$\frac{1}{2} \times 10^{-3}$	0.014	0.815	8.4	0.265	0.313	

N.B. NH4NO4 als N-Quelle.

#### D. III. Kulturen mit Zusatz von CoSO<sub>4</sub>. Aspergillus niger—angestellt 17. Febr.

Geerntet 17. März.

Kulturdauer 28 Tage.

Temperatur 16-20° C.

Gehal	Gehalt in		S	Säure pro 1g.	
Gram Mol.	Gew. %	in g.	Aeidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.267	10.6	0.334	1.251
$\frac{1}{16} \times 10^{-3}$	0.0017	0.393	13.0	0.409	1.041
$\frac{1}{8} \times 10^{-3}$	0.0035	0.561	15.5	0.488	0.870
$\frac{1}{4} \times 10^{-3}$	0.007	0.742	14.8	0.466	0.628
$\frac{1}{2} \times 10^{-3}$	0.014	0.770	10.0	0.315	0.409

N.B.  $\mathrm{NII}_{4}\mathrm{NO}_{3}$  als N-Quelle.

### D. IV. Kulturen mit Zusatz von CoSO<sub>4</sub>. Penicillium glaucum—angestellt 20. März.

Geerntet 30. März.

Kulturdauer 10 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	S	Säure	Säure pro 1g.
Gram Mol.	(i. w. 75	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.108	5.4	0.170	
$\frac{1}{16} \times 10^{-3}$	0.0017	0.186	5.0	0.157	
$\frac{1}{8} \times 10^{-3}$	0.0035	0.225	5.4	0.170	
$\frac{1}{4} \times 10^{-3}$	0.007	0.317	5.9	0.186	
$\frac{1}{2} \times 10^{-3}$	0.014	0.366	5.9	0.186	

N.B. NH<sub>4</sub>NO<sub>3</sub> als N-Quelle.

## D. V. Kulturen mit Zusatz von CoSO<sub>4</sub>. Penicillium g'aucum—angestellt 20. März.

Geerntet 1. April.

Kulturdauer 11 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Sänre pro 1g.
Gram Mol.	Gew. 7	in g.	Acidität	als Oxalsäure nmgerechnet	Pilzsubstanz.
O	0	0.242	4.6	0.145	
$_{16}^{1} \times 10^{-3}$	0.0017	0.469	5.7	0.179	
$\frac{1}{8} \times 10^{-3}$	0.0035	0.354	5.9	0.186	<del></del>
$\frac{1}{4} \times 10^{-3}$	0.007	0.482	5.7	0.178	
$\frac{1}{2} \times 10^{-3}$	0.014	0.772	5.9	0.186	

N.B. NH<sub>4</sub>NO<sub>3</sub> als N-Quelle.

#### D. VI. Kulturen mit Zusatz von CoSO<sub>4</sub>. Penicillium glaucum—angestellt 20. März.

Geerntet 13. April.

Kulturdauer 23 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Aeidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.363	6.4	0.202	
$\frac{1}{16} \times 10^{-3}$	0.0017	0.349	6.2	0.195	
$\frac{1}{8} \times 10^{-3}$	0.0035	0.520	5.9	0.186	
$\frac{1}{4} \times 10^{-3}$	0.007	0.649	5.7	0.179	
$\frac{1}{2} \times 10^{-3}$	0.014	0.289	6.6	0.207	

N.B. NH<sub>4</sub>NO<sub>3</sub> als N-Quelle.

#### E. I. Kulturen mit Zusatz von CuSO<sub>4</sub>. Aspergillus niger—angestellt 21. März.

Geerntet 31. März.

Kulturdauer 10 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	S	äure	Säure pro 1g.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.307	6.2	0.195	0.635
$\frac{1}{16} \times 10^{-3}$	0.0015	0.305	5.2	0.164	0.538
$\frac{1}{8} \times 10^{-3}$	0.003	0.297	5.2	0.164	0.552
$\frac{1}{4} \times 10^{-3}$	0.006	0.311	4.7	0.138	0.444
$\frac{1}{2} \times 10^{-3}$	0.012	0.360	5.1	0.160	0.444

N.B. NH4NO3 als N-Quelle.

# E. II. Kulturen mit Zusatz von CuSO<sub>4</sub>. Aspergillus niger—angestellt 21. März.

Geerntet 5. April.

Kulturdauer 15 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	S	iure	Säure pro 1g.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.273	9.7	0.309	1.121
$\frac{1}{16} \times 10^{-3}$	0.0015	0.307	10.0	0.315	1.003
$\frac{1}{8} \times 10^{-3}$	0.003	0.313	10.3	0.324	1.035
$\frac{1}{4} \times 10^{-3}$	0.006	0.324	10.1	0.318	0.967
$\frac{1}{2} \times 10^{-3}$	0.012	0.345	9.4	0.296	0.858

N.B. NH 4 NO 3 als N-Quelle.

# E. III. Kulturen mit Zusatz von CuSO<sub>4</sub>. Aspergillus niger—angestellt 21. März.

Geerntet 5. April.

Kulturdauer 16 Tage.

Temperatur 16–20° C.

Gehal	Gehalt in		Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgereehnet	Pilzsubstanz.
0	0	0.218	9.9	0.312	1.431
$\frac{1}{16} \times 10^{-3}$	0.0015	0.252	10.1	0.318	1.265
$\frac{1}{8} \times 10^{-3}$	0.003	0.352	9.9	0.312	0.886
$\frac{1}{4} \times 10^{-3}$	0.006	0.358	9.3	0.293	0.818
$\frac{1}{2} \times 10^{-3}$	0.012	0.343	9.6	0.302	0.880

N.B. NH<sub>4</sub>NO<sub>3</sub> N-Quelle.

# F. I. Kulturen mit Zusatz von HgCl<sub>2</sub>. Aspergillus niger—angestellt 1. Febr. '99.

Geerntet 9. Febr.

Kulturdauer 8 Tage.

Temperatur 16–20° C.

Gehalt in		Ernteertrag	S	inre	Säure pro 1g.
Gram Mol,	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
O	0	0.126	12.6	0.397	3.176
$\frac{1}{16} \times 10^{-3}$	0.0017	0.180	13.3	0.419	2.328
$\frac{1}{8} \times 10^{-3}$	0.0034				
$\frac{1}{4} \times 10^{-3}$	0.0067				
$\frac{1}{2} \times 10^{-3}$	0.0135				

N.B. NII  $_4$ NO  $_3$  als N-Quelle, 0.0017  $_7$  gut entwickelt. Sporen braun 0.0034  $_7$  fast keine Entwickelung, 0.0067  $_7$  u. 0.0135  $_7$  keine Entwickelung.

## F. H. Kulturen mit Zusatz von HgCl<sub>z</sub>. Aspergillus niger—angestellt 1. Febr. '99.

Geerntet 9. Febr.

Kulturdauer 8 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.119	10.0	0.315	2.644
$\frac{1}{16} \times 10^{-3}$	0.0017	0.188	12.7	0.400	2.128
$\frac{1}{8} \times 10^{-3}$	0.0034				
$\frac{1}{4} \times 10^{-3}$	0.0067		-		
$\frac{1}{2} \times 10^{-3}$	0.0135				

N.B. NII4NO3 als N-Quelle, Entwickelung wie vorige.

F. III. Kulturen mit Zusatz von HgCl<sub>2</sub>.

Aspergillus niger—angestellt 1. Febr. '99.

Geerntet 9. Febr.

Kulturdauer 8 Tage.

Temperatur 16-20° C.

Gehal	Gehalt in		Säure		Säure pro Ig.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.153	11.3	0.356	2.366
$\frac{1}{16} \times 10^{-3}$	0.0017	0.160	13.7	0.431	2.568
$\frac{1}{8} \times 10^{-3}$	0.0034				
$\frac{1}{4} \times 10^{-3}$	0.0067				
$\frac{1}{2} \times 10^{-3}$	0.0135				

N.B. NH<sub>4</sub>NO<sub>3</sub> als N-Quelle. Entwickelung wie vorige.

F. IV. Kulturen mit Zusatz von HgCl<sub>2</sub>. Penicillium glaucum—angestellt 20. April.

Geerntet 1. Mai.

Kulturdauer 11 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	S	Säure	
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.203	4.5	0.142	
$\frac{1}{16} \times 10^{-3}$	0.0017	0.243	4.7	0.148	
$\frac{1}{8} \times 10^{-3}$	0.0034	0.242	5.1	0.159	
$\frac{1}{4} \times 10^{-3}$	0.0067	0.473	5.1	0.159	
$\frac{1}{2} \times 10^{-3}$	0.0135	0.251	4.9	0.154	

 $\rm N.B.\ NH_4NO_3$  als N-Quelle.

F. V. Kulturen mit Zusatz von HgCl<sub>2</sub>. Penicillium glaucum—angestellt 20. April.

Geerntet 2. Mai.

Kulturdauer 12 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	S	iure	Säure pro 1g.
Gram Mol.	Gew. 7	in g.	Aeidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.222	4.6	0.145	
$\frac{1}{8} \times 10^{-4}$	0.0003	0.282	4.6	0.145	
$\frac{1}{4} \times 10^{-4}$	0.0006	0.264	4.7	0.148	
$\frac{1}{2} \times 10^{-4}$	0.0013	0.273	4.6	0.145	
10-4	0.0027	0.301	4.5	0.142	

N.B. NH<sub>4</sub>NO<sub>3</sub> als N-Quelle.

F. VI. Kulturen mit Zusatz von HgCl<sub>2</sub>. Penicillium glaucum—angestellt 20. April.

Geerntet 1. Mai.

Kulturdauer 11 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	S	Säure	
Gram Mol.	Gew. %	in g.	Aeidität	als Oxalsaure; umger <b>e</b> chnet	Pilzsubstanz.
0	0	0.183	5.0	0.157	
$\frac{1}{8} \times 10^{-4}$	0.0003	0.249	5.5	0.173	
$\frac{1}{4} \times 10^{-4}$	0.0006	0.213	5.9	0.185	
$\frac{1}{2} \times 10^{-4}$	0.0013	0.311	5.5	0.173	
10-4	0.0027	0.246	5.8	0.182	

N.B. NH, NO3 als N-Quelle.

F. VII. Kulturen mit Zusatz von HgCl<sub>2</sub>.

Aspergillus niger—angestellt 30. März.

Geerntet 18. April.

Kulturdauer 18 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Aciditat	als Oxal-äure umgerechnet	Pilzsubstanz.
0	O	0.347	8.8	0.277	0.800
$\frac{1}{8} \times 10^{-4}$	0.0003	0.517	9.6	0.302	0.586
$\frac{1}{4} \times 10^{-4}$	0.0006	0.513	9.4	0.296	0.555
$\frac{1}{2} \times 10^{-1}$	0.0013	0.552	10.9	0.343	0.621
$10^{-1}$	0.0027	0.565	11.3	0.356	0.630

N.B. NII4NO3 als N-Quelle.

F. VIII. Kulturen mit Zusatz von HgCl<sub>2</sub>. Aspergillus niger—angestellt 30. März.

Geerntet 18. April.

Kulturdauer 19 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	5	Säure	Säure pro 1g.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsaure umgerechnet	Pilzsubstanz.
0	0	0.341	8.6	0.271	0.795
$\frac{1}{8} \times 10^{-4}$	0.0003	0.458	9.4	0.296	0.646
$\frac{1}{4} \times 10^{-1}$	0.0006	0.474	9.4	0.296	0.624
$\tfrac{1}{2}\times 10^{-1}$	0.0013	0.630	12.5	0.394	0.625
10-4	0.0027	0.429	10.3	0.324	0.755

N.B. NH<sub>4</sub>NO<sub>3</sub> als N-Quelle.

F. IX. Kulturen mit Zusatz von HgCl<sub>2</sub>
Aspergillus niger—angestellt 30 März.

Geerntet 18. April.

Kulturdauer 19 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Λεidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.261	8.4	0.265	1.015
$\frac{1}{8} \times 10^{-4}$	0.0003	0.355	9.2	0.290	0.816
$\frac{1}{4} \times 10^{-4}$	0.0006	0.380	9.4	0.296	0.779
$\frac{1}{2} \times 10^{-4}$	0.0013	0.509	12.1	0.381	0.742
10-4	0.0027	0.451	11.1	0.350	0.776

N.B. NII, NO3 als N-Quelle.

#### G. I. Kulturen mit Zusatz von LiNO<sub>3</sub>. Aspergillus niger—angestellt 1. April.

Geerntet 18. April.

Kulturdauer 17 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	tiew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	Û	0.300	9.0	0.284	0.946
$\frac{1}{16} \times 10^{-2}$	0.004	0.408	9.4	0.296	0.725
$\frac{1}{8} \times 10^{-2}$	0.008	0.428	8.9	0.283	0.661
$\frac{1}{4} \times 10^{-2}$	0.017	0.348	8.7	0.273	0.784
$\frac{1}{2} \times 10^{-2}$	0.034	0.345	8.6	0.271	0.782

N.B. NH, NO3 als N-Quelle.

## H. I. Kulturen mit Zusatz von NaFl. Aspergillus niger—angestellt 7. Febr. '99.

Geerntet 21. Febr.

Kulturdauer 14 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Aciditat	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.199	8,8	0.277	1.392
$\frac{1}{16} \times 10^{-2}$	0.0025	0.325	9.4	0.296	0.911
$\frac{1}{8} \times 10^{-2}$	0.005	0.312	7.2	0.227	0.727
$\frac{1}{4} \times 10^{-2}$	0.010	0.246	6.1	0.192	0.880
$\frac{1}{2} \times 10^{-2}$	0.021	0.289	6.0	0.189	0.654

N.B. NH4NO3 als N-Quelle.

H. H. Kulturen mit Zusatz von NaFl. Assergillus niger—angestellt 7. Febr. '99.

Geerntet 23. Febr.

Kulturdauer 16 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgereehnet	Pilzsubstanz.
0	0	0.314	10.0	0.315	1.000
$\frac{1}{16} \times 10^{-2}$	0.0025	0.336	10.0	0.315	0.937
$\frac{1}{8} \times 10^{-2}$	0.005	0.385	7.6	0.239	0.621
$\frac{1}{4} \times 10^{-2}$	0.010	0.316	5.9	0.186	0.589
$\frac{1}{2} \times 10^{-2}$	0.021	0.274	5.6	0.176	0.642

N.B. NH<sub>4</sub>NO<sub>3</sub> als N-Quelle.

# H. III. Kulturen mit Zusatz von NaFl. Aspergillus niger—angestellt 7. Febr. '99.

Geerntet 25. Febr.

Kulturdauer 18 Tage.

Temperatur 16-20° C.

Gehalt in		Ernteertrag	Säure		Säure pro 1g.
Gram Mol.	Gew. %	in g.	Acidität	als Oxalsäure umgerechnet	Pilzsubstanz.
0	0	0.270	8.4	0.265	0.982
$\frac{1}{16} \times 10^{-2}$	0.0025	0.280	10.7	0.339	1.207
$\frac{1}{8} \times 10^{-2}$	0.005	0.285	7.7	0.242	0.849
$\frac{1}{4} \times 10^{-2}$	0.010	0.264	6.6	0.208	0.788
$\frac{1}{2} \times 10^{-2}$	0.021	0.265	6.1	0.192	0.725

N.B. NII4NO3 als N-Quelle.

#### INHALT.

- I. Einleitung und Litteratur.
- II. Methodisches.
- III. Vorbemerkungen über die Versuchsobjekte.
- IV. Veränderungen in der Wachsthumsweise und die Correlation zwischen Fortpflanzung und Wachsthum.
- V. Einfluss der Reizstoffe auf die Betriebsstoffwechsel.
- VI. Specielle Besprechungen.
- VII. Schlussbemerkungen und Zusammenfassung der Resultate.

#### TABELLARISCHE ZUSAMMENSTELLUNG.

- I. Versuche mit Algen.
- II. Versuche mit Pilzen.

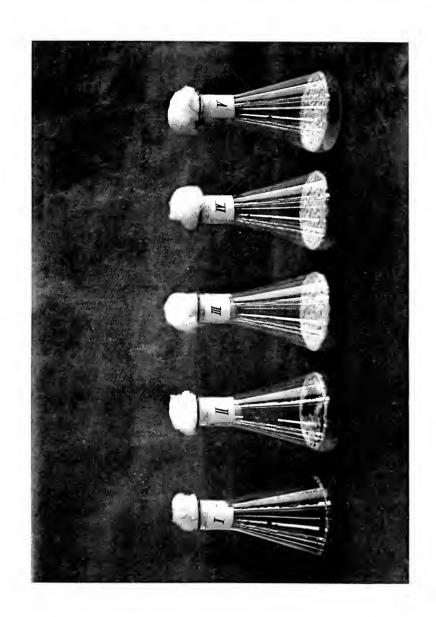
### Erklärung der Tafel XIII.

Kulturen von Aspergillus niger mit und ohne Zusatz von NaFl.

(Photographiert 15 Tage nach der Sporenaussaat.)

- I. Ohne Zusatz; Kontrollkultur.
- II. Mit Zusatz von 0.0025% NaFl.
- III. Mit Zusatz von 0.005%
- IV. Mit Zusatz von 0.010%
- V. Mit Zusatz von 0.021% "

(Für näheres vgl. S. 153 und ferner Tabelle Pilz. H.)





### Ammonium Amidosulphite.

By

Edward Divers and Masataka Ogawa, Imperial University, Tōkyō.

The interaction of such familiar gases as ammonia and sulphur dioxide ceased to attract with any effect the attention of investigators sixty years ago and more. Yet comparatively nothing had then been definitely made out about the nature of the product, and even the few statements concerning it in some of the best treatises on chemistry have but little experimental foundation. The history of the subject is briefly given on p. 193.

Non-union of dry sulphur dioxide and ammonia.

Even when comparatively well-dried, sulphur dioxide and ammonia unite at once and with great energy when brought together; yet they can remain mixed without combining, provided sufficient care has been observed to exclude moisture. It has not been necessary, however, in order to demonstrate this striking phenomenon, to have resort to the elaborate precautions

adopted by Brereton Baker, in his famous experiments upon the non-union of hydrochloric acid and ammonia (J. Ch. Soc., 1894, 65, 611; 1898, 73, 422), and we have only dried the gases during their flow through the tubes. As we were able to dry sulphur dioxide better than ammonia, because common phosphorus pentoxide could be used for the purpose, we have had success in mixing the gases without their combining only by giving this gas precedence. The preparation flask with its tubes having been heated and then kept for a while in the desiccator, was placed in ice and salt while a slow current was sent through it of sulphur dioxide, which had passed through drying-tubes of sulphuric acid and then of phosphorus pentoxide. The outlettube dipped into mercury. Ammonia, dried first by the cold of a freezing-mixture and then by long tubes of freshly fused and crushed potassium hydroxide (but no Stas's mixture), was now also passed into the flask slowly. The result was that the interior of the flask remained clear for some minutes, the mixed gases only combining on their escape through the mercury into the air. But the ammonia having, it is presumed, gradually brought enough moisture with it through passing more rapidly along the tubes than at first, the walls of the flask became suddenly coated with an orange-coloured deposit, while the mercury rose high in the exit tube.

### Proportions in which sulphur dioxide and ammonia combine.

The proportions, in which ammonia and sulphur dioxide combine, or appear to combine together, depend largely upon the extent to which the temperature is allowed to rise, the heat of union being considerable. They vary also according as one or other of the gases is used in excess, unless the temperature is kept very low. But the variation of the proportions and the apparent condensation of additional sulphur dioxide by a sufficiently ammoniated product, that may be observed, are results clearly due to the secondary changes going on (p. 192). The simple union of ammonia and sulphur dioxide, which can be secured by keeping down the temperature by suitable means, especially with the ammonia in excess, is that of two volumes of the former to one of the latter (p. 191). But since this union cannot be made at the ordinary temperature without being immediately followed by a decomposition, in which ammonia is evolved, the union of the two gases can appear to take place in other proportions than the above. It is pretty certain that, by proceeding slowly enough and using strong cooling agencies, secondary action could be almost entirely prevented and the statement just made be verified, even when working with the gases alone. We have not gone very near to getting such a result in this way, but then we have, for good reasons, not striven much to overcome the difficulties. Our experimental work, which will be further on referred to (p. 195), has shown that two much more nearly than one volume of ammonia can be made in this way to unite with one volume of sulphur dioxide, the only proportions which Rose met with in his experiments (p. 193), and that the presence of much ammonium amidosulphite in the product can be established with certainty.

Preparation and analysis of ammonium amidosulphite.

In order to get the primary product of the union of sulphur dioxide with ammonia in its unchanged state, ether was

made use of as the medium of the union, in order to keep the temperature under control. The ether, freed from alcohol and water by sodium, was contained in a small flask, fitted with inlet and outlet tubes, which was to serve, not only for the production of the new substance, but for its isolation and its weighing for analysis. The flask was put in a bath of ice and salt, with the outlet-tube dipping into a trough of mercury, and then the ether was saturated with dried ammonia. Having shut off the ammonia, a very slow current of sulphur dioxide was sent into the solution while the flask was continuously shaken, not only in order to diffuse the heat, but to prevent the product from caking on to the bottom of the flask and shutting in ether. The mouth of the tube conveying the sulphur dioxide soon became filled with a yellow pasty mass (p. 192), and had to be kept open by a platinum rod, manipulated through the rubber tubing above, but the precipitate itself was quite white and powdery. In spite of the external cooling, the heat of combining was sufficient to cause ammonia gas, saturated with ether-vapour, to escape through the mercury sealing the exit-tube, and when this escape became slight, the passage of sulphur dioxide stopped. With the use of about 20 c.c. ether, there had then formed well over a gram of the substance. In order to secure this undecomposed, a second flask was put in connection with the preparationflask, and ammonia again passed to the saturation point. ammoniated ether was decanted off through the connecting-tube into the second flask, which was then detached, the whole operation being carried out in the freezing-mixture. The current of ammonia was renewed over the precipitate in the flask, and continued for hours, until all the ether adhering to the precipitate had been carried away, the flask being all the while still

in the freezing-mixture. There was no other way of completely drying the salt, and even this way was not sufficiently successful when the salt had been allowed to eake together. The ammonia could not be replaced by air or hydrogen for drying the salt, nor could the flask be kept out of the freezing-mixture, so long as ether still moistened the salt, without the latter taking an orange-colour. When dry and in an ammonical atmosphere, the salt is more stable, but cannot long be kept at the ordinary temperature without getting discoloured through decomposition.

Analysis.—The stopper carrying the gas-tubes having been replaced by a plain one, and air allowed to displace most of the ammonia gas, the flask was at once weighed and left for a time inverted with open mouth dipping into 100 c.c., or more, of water in a beaker. When the salt in it had become damp, it was washed into the water, and its very dilute solution distilled with alkali for its ammonia. The residue was divided into two measured portions, one of which was acidified and heated to 150° under pressure for some hours and then redistilled with alkali for additional ammonia, of which only a trace was got (0.001 per cent. of the salt). The other part of the solution was treated with bromine, and next with hydrochloric acid and chlorate, after which barium sulphate was precipitated with the usual precautions. The results of the analysis were:—

	Ammonia	Sulphur diox.
Found:	35.09;	64.91 per cent.
$SO_2(NH_s)_2$ :	34.69;	65.31 "

The slight excess of ammonia indicated is safely attributable to the means taken to preserve the salt till it was analysed.

### Its properties, constitution, and name.

The new salt is white and apparently crystalline, and appears to be slightly volatile in a current of ammonia. very deliquescent and decomposes, losing ammonia, in the air. It dissolves in water, giving out heat and a hissing sound, and if dissolved by ice or enough ice-cold water, furnishes a solution answering all tests for pure ammonium sulphite. In this respect it is quite unlike ammonium amidosulphate or carbamate, since even the latter salt gives at first no precipitate with calcium chloride, which at once precipitates all sulphite from the new When the salt is much decomposed, its solution gives other reactions besides those of a sulphite. In anhydrous alcohol it dissolves freely, evidently as ethyl ammonium sulphite; it is also slightly soluble in dry ether. It soon begins to change and then assumes an orange-colour, even at the common temperature. At 30-35° it decomposes into a liquid and a solid part, both more or less orange-coloured, and into ammonia, the liquid part undergoing further change into solid matters (p. 197)

Constitution.—The salt is more probably an amido-than an imido-compound, NH<sub>4</sub>N(SO<sub>2</sub>NH<sub>4</sub>)<sub>2</sub> (analogue of normal ammonium imidosulphate), because it can be obtained only when the temperature is kept down and the ammonia is in excess. It is still more probably a sulphuryl rather than a thionyl compound, because of its feeble activity as a reducing agent and of its very easy passage into ammonium sulphite or ethyl ammonium sulphite. It has accordingly to be formulated as NH<sub>2</sub>·SO<sub>2</sub>·NH<sub>4</sub>, and not NH<sub>2</sub>·SO·ONH<sub>4</sub>.

Name.—Since the salt represents ammonium sulphite, NH<sub>4</sub>O·SO<sub>2</sub>·NH<sub>4</sub>, in which the ammonoxyl is replaced by amido-

gen, it is properly called ammonium amidosulphite. Berglund's name of amidosulphonate now in use, for amidosulphate is evidently based on a misconception. The name, amidosulphinate, in analogy with amidosulphonate, must be rejected on the same grounds, and because the salt has not the characteristic reducing action and the constitution of sulphinates. It does not seem possible, even were it desirable, to construct a term for the first amide of sulphurous acid that would correspond to that of sulphamic acid, the synonym of amidosulphuric acid.

### Nature of the decomposition by heat of the amidosulphite.

History.—Experiments made earlier than ours on the union of sulphur dioxide with ammonia gave the products of decomposition of ammonium amidosulphite instead of the salt itself. Doebereiner in 1826 (Schw, Jahrb., 17, 120), described the product of the union as a brown-vellow vapour quickly condensing to a bright brown solid mass, which the smallest quantity of water converts into (colourless) ammonium sulphite. Rose published three papers on 'anhydrous sulphite of ammonia' in 1834, 1837, and 1844 (*Pogg. Ann.*, 33, 235; 42, 415; 61, 397), in the second correcting statements made in the first, and modifying in the third the views he had expressed in the earlier papers. The outcome was that he had ascertained that the product of the union is always one and the same single substance, in whatever proportions the dry gases are taken; that it is composed of equal volumes of the gases, is either yellowish-red and smeary, or red crystalline, very deliquescent and very soluble in water without evolving ammonia; that it yields a neutral solution, which is at first yellowish but soon, becomes

colourless, and gives, when recently prepared, the reactions mainly of a mixture of ammonium sulphate and trithionate, but to a small extent those of a sulphite also; and, lastly, that when the solution is of certain concentration it gives a transient reddish coloration with hydrochloric acid.

Forchhammer (Compt. rend., 1837, 5, 395) found that, besides the orange-coloured substance, crystals of ammonium sulphate are produced by the union of the gases, which can sometimes be seen apart from the other product in some spots of the mass, though often indistinguishably mixed up with it. (That the crystals observed in the product were those of sulphate, could only have been a supposition of Forchhammer's). The mass when moistened is alkaline and evolves ammonia, yielding otherwise the reactions recorded by Rose. Absolute alcohol dissolves out of it a substance which takes a rose colour, soon disappearing. Indirectly, he represented the mass to be derived from two mols. ammonia to one mol. sulphur dioxide, as did also Doeberenier.

The views advanced as to the nature of the orange body have been, that it is a compound of ammonia with an isomer of sulphurous anhydride, which changes at once with water into ammonium sulphate and trithionate, just as ammonium pyrosulphite slowly changes in hot solution (Rose); that it is amidogen sulphide,  $S(NH_2)_2$ , mixed with ammonium sulphate (Forchhammer); that it is, partly, thionamic acid,  $NH_2$ :SO·OH, partly, ammonium thionamate, both volatile, being its colour due to impurity (H. Watts); and that it is ammonium pyrothionamate,  $NH_2$ :S<sub>2</sub>O<sub>4</sub>·NH<sub>4</sub> (Joergenssen).

Interaction of the gases.—We have repeated Rose's experiments of measuring over mercury the volumes of the gases which

interact, in which he found that always equal volumes combine, whichever gas may be taken in excess. The results somewhat approached this when no steps were taken to restrain the rise in temperature due to the union of the gases; but when the gastube was immersed in a cooling-mixture and the ammonia was in excess, the volume of this gas consumed was much greater than that of the sulphur dioxide. This method of investigating the matter is, however, inapplicable, because the ammonium amidosulphite, which is formed, partly decomposes with free evolution of ammonia. By letting the dried gases come together in a vessel agitated in a freezing-mixture and keeping the ammonia in excess, a solid mass is obtained which consists largely of the amidosulphite, behaving as such in water, though mixed with other substances, and quantitative analysis of which shows that much more than three mols. ammonia to two mols. sulphur dioxide have gone to its formation. If, instead of examining it at once, it is kept for a long time in a gentle current of dry nitrogen or hydrogen, at a temperature of 30° to 35°, it no longer contains amidosulphite or gives any sulphite to water, and contains not much more than one atom of nitrogen to one of sulphur. Thus, Rose's results are explained and, at the same time, shown to be of no direct significance.

Products of the decomposition.—Both Rose and Forchhammer found ammonium sulphate to be a principal constituent of the product of the interaction of the gases. A sufficiently high temperature having been reached, this will have been the case; furthermore, the solution of the even less heated product slowly becomes acid and full of sulphate. But when the temperature has not been allowed to exceed 30°, or even 40°, the quantity of sulphate in the product is so small that it may almost be dis-

regarded. Along with sulphate, trithionate was considered by Rose to make up most of the product, for the aqueous solution of the mass always gives a strong reaction with silver nitrate which might be that of trithionate and, in the case of his product, gave other reactions of a trithionate. But when the product has been carefully prepared and is free from amidosulphite, its solution gives the silver reaction without the others belonging to a trithionate. Thus, the solution may be acidified and left for hours without yielding more than mere traces of sulphur dioxide and sulphur; to get these in quantity, the solution had to be strongly heated under pressure. Besides this, the absence of sulphate in the solution is of itself almost enough to disprove the production of trithionate, since, as Rose himself represented, sulphate and trithionate are complementary products of the decomposition.

Heating pure ammonium amidosulphite gives the same results as heating the coloured product of the union of sulphur dioxide and ammonia as gases. Rose's assertion that the product is a single substance, even in appearance, is certainly incorrect, according to our experience. By the union of the gases in a receiver kept well cooled, the product is deposited as a soft, waxy, yellow coating on the walls of the vessel and on the gas tubes. Its colour varies in different parts from nearly white to orange-red somewhat irregularly but generally so as to be whiter near where the ammonia enters, the whiteness not being due to moisture in the gases, as Rose assumed. When the product gets to 30-35°, whether by its own heating or by external heat, it is decomposed at first into an obscurely crystalline white solid and a much smaller quantity of a coloured, effervescing liquid, partly draining to the bottom of the vessel; after a time all becomes

solid again and tenaciously adherent to the glass. When pure ammonium amidosulphite is similarly heated in a dry inactive gas, it colours, softens, shrinks together, vesiculates, gives out ammonia, and becomes a mass like that derived directly from the union of the gases. With very gradual heating, the temporarily liquid product is much less coloured than in the other case, its colour being evidently caused by the presence of a red matter dissolved in it, which gives indications of being volatile.

This orange-red substance is never formed in more than very small quantity. It gives a yellow colour to the aqueous solution of the whole product, which, however, slowly fades away. Alcohol, carbon bisulphide, and other menstrua dissolve it out from the salts, leaving them white; but the solutions are not pure. The yellow solution in water or alcohol takes a transient pink colour when mixed with dilute hydrochloric acid, and the alcoholic solution an indigo-blue colour with concentrated ammonia. The residue left on evaporating the carbon-bisulphide-solution becomes explosive when heated above 150°, and may then have become nitrogen sulphide, but before being heated it is not this substance.

Except the very little sulphate already mentioned, there is no as-yet known substance present in the residue of the decomposition of the amidosulphite by a gentle heat, so far as we can discover. Alcohol of 90 per cent. dissolves out something, but only very sparingly. By evaporation of the solution in a vacuum desiccator, a very deliquescent salt is obtained in crystals, having a composition that may be expressed by 9NH<sub>3</sub>, 8SO<sub>2</sub>, assuming the presence of 2.5 per cent. moisture. The composition of the whole crude residue does not differ much from this. The alco-

holic solution, cooled and charged with ammonia, gives minute scaly crystals in small quantity. This substance, dried in a current of ammonia, has a composition expressed by  $(NH_3)_3S_2O_3$ , and dried in the sulphuric-acid desiccator, that of  $(NH_3)_2S_2O_3$ . These three substances all give the silver-nitrate reaction of the aqueous solution of the whole residue, and on boiling with dilute hydrochloric acid give very little sulphur and no sulphur dioxide. At higher temperatures, whether dry or in solution, they yield sulphur, sulphur dioxide, and sulphate. Two potassium derivatives of these salts have also been prepared. Neither the crude residue nor any of the above substances yields all its nitrogen as ammonia when distilled with alkali, unless it has been first heated with hydrochloric acid under pressure.

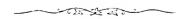
From the mother-liquor of the above mentioned S<sub>2</sub>O<sub>3</sub> salt a substance was got which in composition and behaviour appeared to be sulphamide a little impure. Neither sulphamide nor amidosulphate can be found in the fresh aqueous solution of the whole residue, but, by heating the solid residue itself to a higher temperature, imidosulphate is obtained in considerable quantity, besides sulphur and sulphate, and imidosulphate is a known product of first heating and then dissolving in water, either amidosulphate or sulphamide. A proof-spirit extract and also a wood-spirit extract of the residue yield ammonium amidosulphate on evaporation, no doubt generated by hydration. An aqueons solution of the less heated residue, treated with excess of barium acetate and filtered, gave barium thiosulphate in crystals, on evaporating it over the water-bath.

During the heating of ammonium amidosulphite at a temperature of 30° to 35°, besides much ammonia, small quantities of water and of sulphur dioxide are evolved, the former mainly

in the early stage and the latter in the late stage of the decomposition. This remarkable production of water, though always evident, was fully established by cooling the escaping gases and testing the water thus collected. The presence of sulphur dioxide later in the operation was shown by the gases fuming on their escape into the air and then forming a small white deposit, slowly turning orange, and reacting as ammonium pyrosulphite. In the interaction of sulphur dioxide with ammonia, and in the decomposition of the amidosulphite, no liberation of nitrogen could ever be discovered.

To sum up the results of our incomplete work upon the decomposition of ammonium amidosulphite by a graduated and gentle heat, ammonia and a residue consisting of a substance (or substances), which behaves as a thio-amido-sulphonic compound, are the principal products; in much less quantities, water and an orange-red substance are also produced, and, generally if not always, a very little sulphate; and, as secondary products, apparently sulphamide and certainly amidosulphate and thiosulphate are obtainable, as well as imidosulphate, sulphur, and much sulphate. It seems of interest to point out that we here record the first production known of amidosulphate from ammonia and sulphur dioxide, which, hitherto, has been derived either from ammonia and sulphur trioxide or from a nitrite and sulphur dioxide.

We hope in a future paper to be able to report the completion of this investigation.





## Products of heating Ammonium Sulphites, Thiosulphate, and Trithionate.

By

### Edward Divers and Masataka Ogawa,

Imperial University, Tōkyō.

What has been published upon the effects of heating ammonium sulphites and thiosulphate is but little in accordance with the results of experiments we have had to make upon these salts and upon the hardly known trithionate, in connection with an investigation of the decomposition by heat of ammonium amidosulphite. We therefore make known what we have ascertained.

### Preparation of the salts used.

Ammonium sulphite, (NH<sub>4</sub>)<sub>2</sub> SO<sub>3</sub>, OH<sub>2</sub>.—Statements are conflicting as to whether this salt can be got from its solution by evaporation (Muspratt, *Phil. Mag.*, 1847, iii, 30, 414; Marignac, *Jahresb.*, 1857, 17; Forerand, *Compt. rend.*, 1885, 100, 245; Hartog, *Compt. rend.*, 1887, 104, 1793; Roehrig, *J. pr. Ch.*, 1888, 37, 227). We find that a concentrated solution, charged with

ammonia, can be quite successfully made to deposit the salt by cold evaporation in a potash desiccator, but to get such a solution the moderately strong solution of ammonia, which must be used, has to be kept very cold while passing in the sulphur dioxide. Dilute solutions fail to yield the salt on evaporation because too much of it suffers decomposition. Much better than evaporating is to take advantage of the lessened solubility of the salt in presence of much ammonia. Ammonia solution, sp. gr. 0.895, containing therefore about 28 grm. ammonia in 100 c.c., is to be treated in a flask with sulphur dioxide, while it is kept in motion in a mixture of ice and salt, and with the tube conveying the sulphur dioxide not dipping into the solution. The formation of a very little orange-coloured matter in the neck of the flask cannot be avoided, but this can be easily removed afterwards. When the solution has become thick with crystals, no more sulphur dioxide is to be added, although very much ammonia still remains. Even at the common temperature the crystals do not sensibly dissolve in presence of this ammonia. The salt, drained on a tile under close cover, can be dried either by filter paper or by only short exposure in the desiccator over potassium hydroxide or carbonate, salted just before with ammonium chloride. It is equivalent in quantity to about onefourth of the ammonia taken. By long exposure in a dried atmosphere the salt becomes anhydrous without loss of ammonia. Exposed to the air, it is apparently deliquescent but in reality it evolves ammonia and thus becomes the very deliquescent pyrosulphite.

Anhydrous ammonium sulphite is readily obtained from the hydrated salt by long enough exposure in the desiceator; it is very hygroscopic.

Ammonium pyrosulphite, (NH<sub>4</sub>)<sub>2</sub>S<sub>2</sub>O<sub>5</sub>.—When, in the process just given for preparing the normal sulphite, the passage of sulphur dioxide is not stopped when the solution is full of crystals, these gradually dissolve up and the solution becomes greenishyellow. Then, as it gets charged with sulphur dioxide, in the cooling mixture, the pyrosulphite crystallises out from it, in quantity equivalent to a little over one-fifth of the ammonia taken, being thrown out of solution by the sulphur dioxide. The salt can be obtained dry and pure in the same way as the normal sulphite, except that sulphuric acid, to which a little solid alkali sulphite has been added, is used in the desiccator, though it is very deliquescent and changeable when not carefully preserved from moisture. This salt is also easily obtainable by evaporating its aqueous solution, but hardly free from sulphate, and not without some decomposition, through loss of sulphur dioxide and through oxidation. It is much more soluble than the normal sulphite.

Ammonium thiosulphate.—An old solution of calcium thiosulphate, obtained by boiling lime and sulphur together in water and leaving the solution until much of the pentasulphide had been oxidised by the air, was decanted from insoluble matters, mixed with ammonium carbonate in some excess, filtered, and then freely exposed to the air for some time at 50-60°. In this way a very concentrated solution of ammonium thiosulphate was obtained, free from sulphate and other salts. The solution of this very soluble salt was then dried up to a crystalline mass in the desiccator. The well-dried crystals have been found by Lock and Kluess (Ber., 1889, 22, 3099) to be anhydrous.

Ammonium trithionate.—This salt has apparently not hitherto been prepared by any one. Being exceedingly soluble in water, it cannot be prepared by Plessy's excellent process for the potassium salt (Ann. Ch. Phys., 1844, iii, II, 182), or by its slight modification by Hertlein (Z. phys. Ch., 1896, I9, 287). We therefore made the pure potassium salt by Plessy's method, precipitated the potassium from it by hydrofluosilicic acid, neutralised quickly with ammonia, and precipitated the ammonium trithionate by absolute alcohol and dried it in the desiccator. This very deliquescent and changeable salt cannot be kept long in good condition, but it was used by us when freshly prepared and while still almost free from sulphate.

## Effects of heating the salts.

The process.—The salts were heated in an oil-bath, in a subliming vessel consisting of a test-tube, 15 cm. long and about 15 mm. in internal diameter. The tube was closed by a caoutchouc stopper, and a very slow current of dried nitrogen through the tube was maintained during the heating and cooling. The salt, usually about 4 grm., was contained in an open slender bottle, about 6 cm. long, having a platinum wire attached to it for lowering it into and lifting it out of the subliming tube. The tube was immersed in the oil to the level of the mouth of the bottle inside, so as to cause all dry sublimates to collect in the tube above this level. When, as in the ease of the hydrated normal sulphite, the heating was divided into stages, the bottle was transferred between these to a second subliming-tube. The heating of the oil was conducted very slowly, so that the temperatures mentioned which were those of the oil, may be accepted as being very nearly those of the salts at the time.

In describing the effects of heating them, the salts are taken

in the inverse order of that followed above, in accordance with usage. This is done because of the nature of the products.

Ammonium trithionate.—This salt is hardly affected until the temperature is above 150°, and at 160-170° it steadily decomposes into sulphur dioxide and a residue of ammonium sulphate and unfused sulphur. The non-fusion of the sulphur is remarkable and only to be referred to the presence of minute quantitities of impurities. It all dissolved readily in carbon bisulphide, and crystallised out on evaporating the solvent.

It can hardly be doubted but that ammonium tetrathionate (and pentathionate, if it can exist) would decompose in the same way as trithionate. Ammonium hyposulphate (dithionate) has been shown by Heeren (Pogg., 1826, 7, 55), and more definitely by Kluess (Ann., 1888, 246, 194) to first become anhydrous, if not already so when heated, and then to decompose at about 130° into sulphur dioxide and a residue of ammonium sulphate.

Ammonium thiosulphate.—Zeise, in 1824 (Gm. Hbk) found this salt to be converted by heat into water, ammonia, and a sublimate of sulphur, much thiosulphate again and sulphite, and a little sulphate. This result must have been obtained by rough heating. A much more weighty statement is that made by Spring (Ber., 1874, 7, 1159), namely, that the dry salt can be sublimed unchanged, intermediate dissociation being admitted. We have found it to decompose very slowly at 150°, the main products being a sublimate of anhydrous normal sulphite and a residue of sulphur unfused, as in the case of the trithionate. But, also very small quantities of hydrogen sulphide and ammonia passed off in the current of nitrogen, and the sublimate contained a very little of a salt having some of the properties of trithionate and which did not strike the violet colour with

ferric chloride given by a thiosulphate. Analysis of the sublimate and of that part of the salt which remained mixed with the sulphur when the progress of the decomposition was arrested after only half of it had been decomposed, gave results that showed the former to be essentially anhydrous normal sulphite, and the latter unchanged thiosulphate:—

	$\mathbf{A}\mathbf{m}\mathbf{m}\mathbf{o}\mathbf{n}\mathbf{i}\mathbf{a}$	Sulphur
$(\mathrm{NH_4})_2\mathrm{SO}_3$	29.31	27.59 per cent.
Sublimate	27.54	27.55 ,,
$({ m NH_4})_2 { m S}_2 { m O}_3$	22.27	43.24 ,,
Residue	20.69	42.31 ,,

The main decomposition of the thiosulphate is in full agreement with the relation of thiosulphates to sulphites. Very interesting is the production of a little ammonia and hydrogen sulphide, in connection with the relation of trithionate to thiosulphate as its thio-anhydride (Spring):—2(NH<sub>1</sub>)<sub>2</sub>S<sub>2</sub>O<sub>3</sub>=2NH<sub>3</sub>+SH<sub>2</sub>+(NH<sub>1</sub>)<sub>2</sub>S<sub>3</sub>O<sub>6</sub>. When ammonium thiosulphate is rapidly and more strongly heated, ammonia is lost and sulphur sublimes; then as a matter of course and of no significance, thiosulphate and even trithionate are produced on adding water to the mixed sublimates.

Ammonium pyrosulphite.—We did not get this exceedingly deliquescent salt into the tube ready for heating before it had condensed some moisture, and to this we attribute part of the results obtained. Change went on slowly in the salt at 130° and somewhat faster at 150°. At first there was little else than a slight but steady evolution of sulphur dioxide, and this continued though very feebly, to the end and while a sublimate forming. The sublimate was pyrosulphite in one experiment; in another, it was this salt mixed with a very little anhydrous sulphite. But there was a

considerable residue, more than one-third of the weight of the salt taken, consisting of sulphate, trithionate, sulphur, and apparently some tetrathionate. There was no sulphite or thiosulphate. The tetrathionate, the sulphur, and the sulphur dioxide were very probably derived from decomposition of trithionate by moisture. From a consideration of the results it seems almost necessary to assume that perfectly dry pyrosulphite sublimes unchanged (with no doubt intermediate dissociation), and that the presence of a little moisture causes it to decompose partly into sulphate and trithionate.

Anhydrous ammonium sulphite volatilises at about 150°, yielding a sublimate of the same salt, or rather, a pseudosublimate, for the salt surely dissociates when heated.

Hydrated ammonium sulphite.—According to Muspratt, this salt all volatilises when heated, no sulphate being produced, and yields water, then much ammonia, and finally a sublimate which, judging, from its properties, is ammonium pyrosulphite. observed the following effects of gradually heating it in a very slow current of dried nitrogen. At about 90°, the salt moistened and escape of ammonia became quite evident, and at a little above 100° distillation of water also took place; both water and ammonia continued to escape in noticeable quantities for  $2^{1}/_{2}$  hours longer, when the temperature for some time had been 120°; up to this, a very little sublimate only had formed and matters were now almost at a standstill. The quantity of the salt heated was about 4 grm., and this had now lost one-fifth of its weight, the residue having the composition expressed by  $(NH_3)_{10}(SO_2)_6(OH_2)_7$ , equivalent to a mixture or combination of the three salts, hydrated sulphite (39.4%), anhydrous sulphite (34.1%), and pyrosulphite (26.5%), dividing equally among themselves the sulphur dioxide. Some repetitions of the experiment gave almost the same results. Calculation and the results of one experiment gave the following numbers:—

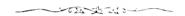
	Ammonia	Sulphur dioxide	
$(NH_3)_{10}(SO_2)_6(OH_2)_7$	25.00	56.47 per cent.	
Found	24.65	56.20 ,,	

If, in the formation of this complex, no longer losing material quantities of ammonia and water, only these products had been given off, the residue should have been  $84^{1}/_{2}$  per cent. of the hydrated normal sulphite, whereas it proved to be little more than 79 per cent., in consequence of volatitisation of some of the (dissociated) salt, made manifest by the production of a little sublimate.

After renewing the heating in a fresh subliming-tube, allowing the temperature to rise slowly from 120° to 150°, the residue had almost all disappeared in two hours, while an abundant dry sublimate had deposited. For some time during this heating, sulphur dioxide steadily escaped, but practically ceased to do so long before sublimation was finished. The residue left when sulphur dioxide was no longer coming off, proved on analysis to be normal sulphite again, but only half hydrated,  $2(NH_4)_2SO_3$ ,  $OH_2$ . The sublimate, also, now and at the finish, consisted of normal sulphite, apparently anhydrous though found to be a little hydrated because it is very hygroscopic and had unavoidably some exposure to the air while it was being scarped out of the tube into the weighing bottle.

Hydrated ammonium sulphite, therefore, becomes by gradual heating to 120° converted one-third into the anhydrous salt, and one-third into pyrosulphite, by loss of water and ammonia; and then the nearly stable complex of these salts with

the other third of the original salt becomes converted into the nearly anhydrous normal sulphite, between 120° and 150°, sulphur dioxide and water escaping. The presence of water is essential to the occurrence of both changes; dry ammonium pyrosulphite partly sublimes as such at 150° and partly changes into sulphate and trithionate, as already described. Heating in the open tube, and more rapidly, Muspratt's results will be got, for then weter is more quickly expelled, and some pyrosulphite can deposit as a sublimate.



# Potassium Nitrito-hydroximidosulphates and the Non-existence of Dihydroxylamine Derivatives.

By

Edward Divers, M. D., D. Sc., F. R. S., Emeritus Prof.,

and

Tamemasa Haga, D. Sc., F. C. S., Professor, Tōkyō Imperial University.

Like potassium nitrate (this Journal, 7, 56), potassium nitrite forms double salts with the potassium hydroximido-sulphates (sulphonates), the non-recognition of whose existence has allowed mistaken notions to arise about the nature and the products of the sulphonation of nitrous acid.

Potassium nitrite and 2/3 normal hydroximidosulphate, KNO<sub>2</sub>, HON(SO<sub>3</sub>K)<sub>2</sub>.—The sparing solubility of 2/3 normal potassium hydroximidosulphate in water is hardly affected by the presence of potassium nitrite and when a sufficient quantity of the salt has been dissolved by heat it crystallises out again almost pure on cooling the hot solution, even though the water has also dissolved in it as much as one-sixth of its weight of the nitrite. When the solution of the nitrite is stronger than this there crystallises

out instead of the hydroximidosulphate itself a combination of it with a molecule of the nitrite. The same double salt is also formed in the cold when the hydroximidosulphate is triturated and digested with such a solution of the nitrite. Precautions being taken against the hydrolysis of the unstable hydroximidosulphate this salt can be dissolved at 70° in as little as 3.8 times its weight of a 22 per cent. solution of nitrite and by cooling the solution the double salt be got in crystals in quantity equivalent to about 12/13 of that of the hydroximidosulphate.

While the hydroximidosulphate itself crystallises in hard rhombic prisms with 20H2, its compound with the nitrite is in silky asbestus-like fibres which are anhydrous. The compound salt is also not deliquescent although potassium nitrite alone is very deliquescent. There is nothing else in its properties whereby to distinguish it from a mixture of its component salts. can be recrystallised from a hot solution of potassium nitrite of a strength of 10 per cent. or more nitrite. It is neutral to litmus and very soluble in water but its solution soon deposits erystals of the 2/3 normal potassium hydroximidosulphate unless it is very dilute. In any case the hydroximidosulphate can be precipitated and thus separated from the nitrite by the addition of barium hydroxide. Like a simple hydroximidosulphate (this Journal, 7, 40), the solid salt digested with a highly concentrated solution of potassium hydroxide is converted into sulphite and nitrite. When acidified its solution becomes yellowish for a short time and then effervesces from the escape of nitrous oxide, a result of the hydroximidosulphate being a sulphonated hydroxylamine, for hydroxylamine and nitrous acid decompose together into nitrous acid and water, the other product in the present case being potassium acid sulphate only. It decomposes

explosively when heated—more so than does the hydroximidosulphate by itself—giving off almost colourless gases and white fumes, just as might be expected and just as does a dry mixture of its constituent salts in corresponding proportions or a mixture of nitrite with a little sulphite.

The compound salt can be purified from other salts or from alkali when these are present by recrystallising from strong enough potassium nitrite solution. But from its own mother-liquor it can be separated only by draining on the tile and not by washing. Such draining however is very effective because of the felted fibrous form of the salt, its non-deliquescent nature, and the hygroscopic character of a solution of potassium nitrite. The analysis of the salt was made in the usual way described in our previous papers on hydroximidosulphates and other sulphonated-nitrite derivatives. By boiling its solution with an acid most of its sulphur appears as ordinary sulphate, but not quite all; so that in estimating the sulphur the solution must be hydrolysed for some hours at 150° under pressure. The results of analysis were:—

	Potassium	Sulphur
Found,	33.14	17.95 per cent.
$K_2HN_2S_2O_2$ ,	33.10	18.06 .,

There are other ways in which the potassium nitrito-2 3 normal hydroximidosulphate may be formed all consisting essentially in producing the hydroximidosulphate by sulphonating a small portion of the potassium nitrite in a concentrated solution. Thus the following mode of working will give good results with certainty but it may be widely deviated from with due consideration and precaution provided only that a concentrated solution

of nitrite be employed. Potassium nitrite, 30 grams; potassium hydroxide, 10 grams; water, 50 to 100 grams are to receive a current of sulphur dioxide freely until crystals begin to form, the containing flask being all the time agitated in a cooling bath of ice and brine. The sulphur dioxide is now to be entered more slowly for some time longer and then stopped. After letting the flask stand for half an hour the solution should be full of the desired salt which is then drained dry on the tile. Its mother-liquor is alkaline to litmus but not to rosolic acid (presence of sulphite, absence of alkali); the well-drained salt itself is only faintly alkaline to litmus, if at all so. The double salt is also produced when to an ice-cold nearly saturated solution of potassium nitrite a similar solution of potassium pyrosulphite is very slowly added until crystallisation begins after which the solution is allowed to stand for some time. Thus prepared, the compound salt is liable to be contaminated with a little nitrilosulphate and sulphite. The experiment just described was made first by Raschig but he attached to it a significance unlike that here presented. Discussion of his views will be found towards the end of this paper.

There is yet another way in which this potassium nitritohydroximidosulphate can be produced which it is of interest to mention because it illustrates the decomposibility of potassium 5/6 normal hydroximidosulphate into the normal and 2/3 normal salts. While the 2/3 normal salt dissolved in 16 per cent. or richer solution of the nitrite crystallises out only in combination with nitrite, the 5/6 normal salt can be dissolved in a nitrite solution of even 50 per cent. and yet for the most part crystallise out again uncombined. But generally with this strength of nitrite solution a little fluffy or cotton-like lustreless matter also

separates. If now to this fluffy matter suspended in its cold mother-liquor carefully decanted from every particle of the crystals of the 5/6 normal salt a hot solution of this 5/6 normal salt in 50 or even 40 per cent. nitrite be poured in, a relatively large quantity of the fluffy matter is obtained and not the hard prisms of the 5/6 normal salt. Under the microscope the fluffy matter proves to be crystalline and when drained on the tile it exhibits a silvery lustre while on analysis it proves to be the nitrito-2/3 normal hydroximidosulphate only slightly impure from the presence of a little 5/6 normal hydroximidosulphate and nitrite. Thus in place of the potassium 33.10 and sulphur 18.06 per cent. we found in it 33.79 and 18.35 respectively, together with an alkalinity equal to 1.09 per cent. potassium. Dissolved up in hot 12 per cent. nitrite solution it recrystallises as the pure double salt. It is thus apparent that in a very concentrated solution of nitrite containing the 5/6 normal salt dissolved there is unstable equilibrium between the tendency to yield HON (SO<sub>3</sub> K)<sub>2</sub>, KON(SO<sub>3</sub>K)<sub>2</sub>,OH<sub>2</sub> again and that to form HON (SO<sub>3</sub>K)<sub>2</sub>, KONO.

Sodium nitrite forms a compound with sodium 2/3 normal hydroximidosulphate which has not been further examined principally because of its high solubility in sodium-nitrite solution.

Potassium nitrite and normal hydroximidosulphate KNO<sub>2</sub>,  $2\text{KON(SO}_3\text{K)}_2$ ,  $4\text{OH}_2$ .—This compound salt is only obtainable from a strongly alkaline solution. For when the normal hydroximidosulphate is dissolved in a hot concentrated solution of the nitrite only the 5/6 normal hydroximidosulphate crystallises out on cooling just as it would do in the absence of nitrite. In order to crystallise out either the normal hydroximidosulphate (this Journal, 7, 30) or its combination with nitrite free alkali

must be present in some quantity in the solution. The presence of too much alkali causes a little of it to separate with the normal salt, taking the place apparently of the water of crystallisation of this salt (this Journal 7, 52), and similarly to separate with the normal salt in its combination with nitrite, then also seeming to lessen the capacity of the normal salt to take up nitrite. The double salt is readily obtained by dissolving normal hydroximidosulphate nearly to saturation in a hot (70°) solution consisting of 33-66 parts nitrite and 3-5 parts hydroxide to 100 parts water and cooling. Usually it forms lustrous silky fibres like those of the 2/3 normal double salt but radiating from points to form voluminous soft spherical masses. When the solution is more strongly alkaline the double salt separates as nearly opaque spherical granules with sometimes long fibres growing out from them. Under the microscope these granules are seen to have also a radiating fibrous texture and to represent the soft voluminous spheres highly condensed. Probably these always begin their growth from a minute granular nucleus. The double salt can only be purified for analysis by pressing it on the porous tile, when the soft spheres become a felted lustrous cake and the hard white granules crumble down like masses of wax. Analysis of the two forms has given us the following results:--

	Potssm.	Alk. potssm.	Sulphur
Silky; found,	35.21	9.92	16.23 per cent.
$K_7N_5S_4O_{16}$ , 4.4 $OH_2$ ,	35.14	10.04	16.43 ,,
Granular; found,	33,99	9.20	15.90 "
$K_7N_3S_4O_{16}$ , $6OH_2$ ,	33.89	9.68	15.85 .,

The varying amount of water is only the recurrence of what we

have recorded concerning the normal potassium hydroximidosulphate by itself. The double salt is exceedingly alkaline, its alkalinity we estimated by means of decinormal acid and litmus.

Like the previously described double salt it is but little soluble in concentrated nitrite solution and freely soluble in water which decomposes it into its constituent salts and also decomposes one of these, the normal hydroximidosulphate, into alkali and crystals of the 5/6 normal salt. When heated it decomposes suddenly but gently and without fusing or scattering, and evolves slight red fumes only. It was by this behaviour quite distinguishable from the 2/3 normal double salt and also from any other hydroximidosulphate which, simple or combined with nitrite, contained less than its  $K_7$  to  $S_4$ . By dissolving the nitrito-2/3 normal hydroximidosulphate in a hot concentrated solution of nitrite containing sufficient alkali the nitrito-normal hydroximidosulphate can be readily obtained by cooling the solution.

Potassium nitrite and potassium 5/6 normal hydroximidosulphate.—We have obtained three compounds of the 5/6 normal
salt with nitrite, one being 7KNO<sub>2</sub>, 2HK<sub>5</sub>(NS<sub>2</sub>O<sub>7</sub>)<sub>2</sub>, 3OH<sub>2</sub>. By
using an almost saturated solution of potassium nitrite containing a little potassium hydroxide and dissolving in it by heat
the 5/6 normal hydroximidosulphate there is obtained a compound
in minute fibrous crystals very lustrous when dry and decomposed
by water but recrystallisable from a saturated nitrite solution.
The same compound salt can be obtained also by dissolving the
nitrito-normal hydroximidosulphate in hot almost saturated solution of nitrite.

Heated it proves to be mildly explosive. Its composition approaches that indicated by the formula given above. For

analysis it was only air-dried on the tile; in the desiccator it would probably have lost its 3 per cent. of water (=3OH<sub>2</sub>) and then approached in composition Fremy's *sulphazite*.

	Potssm.	Sulphur	Alk. Potssm.
Original salt, found,	36.81	14.35	4.80 per cent.
Recrystallised, ,,	36.68	14.47	4.51 ,,
$K_{17}H_2N_{11}S_8O_{42}$ , $3OH_2$ ,	36.87	14.20	4.34 ,,

A second double salt,  $3KNO_2$ ,  $K_5H$  ( $NS_2O_7$ )<sub>2</sub>,  $OH_2$ , was got by dissolving one mol. 5/6 normal salt and 1.4 mol. potassium hydroxide in a hot 65 per cent. nitrite solution and cooling. In appearance it resembled the other compound salt. Its analysis gave:—

	Potssm.	Sulphur	Alk. potssm.
Found,	36.17	15.07	4.51 per cent.
Calc.,	36.81	15.06	4.61 ,,

A third double salt, anhydrous, 7KNO<sub>2</sub>, 3K<sub>5</sub>H(NS<sub>2</sub>O<sub>7</sub>)<sub>2</sub>, was not prepared synthetically but by treating an almost saturated solution of the nitrite with alkali and sulphur dioxide, and adding alkali again after the sulphonation, imitating a process of Fremy's. Then, filtering the heated solution from much crystalline 5/6 normal hydroximidosulphate mixed with a little of its combination with nitrite, we got the mother-liquor, when quite cold, almost filled with tiny prisms of a compound answering to the above formula:—

	Potssm.	Sulphur	Alk. potssm.
Found,	36.94	16.37	4.96 per cent.
Calc.,	36.99	16.51	5.05 ,,

This salt was quickly resolved by water into nitrite and

crystals of the very sparingly soluble 5/6 normal hydroximidosulphate.

The varying proportions in which potassium nitrite and the 5/6 normal hydroximidosulphate unite would possess but little interest were it not for the fact that they have evidently been severally met with and taken to be salts of specific constitution by Fremy and by Raschig.

#### Non-existence of Dihydroxylaminesulphonates.

Fremy believed in the existence of less sulphonated derivatives of potassium nitrite than his sulphazite (see next paper) itself less sulphonated than his *sulphazotates* (hydroximidosulphates) and attributed his failure to find them to the fact of their possessing exceedingly high solubility. Claus held much the same views and believed that by adding to an aqueous solution of potassium nitrite an alcoholic solution of sulphur dioxide in not too large a quantity he had obtained an impure crystallisation of a salt, ON SO<sub>3</sub>K (Ber. 1871, 4, 508): he did not prove this to be the case, but what he did publish about his product is sufficient to show us that he had got the compound of potassium nitrite with 2/3 normal hydroximidosulphate we have described in this paper. A repetition of his experiment gave us this double salt together with much ethyl nitrite. Raschig regarded Claus's preparation as essentially the same as one of his own salts to which he gave the constitution of basic dihydroxylamine sulphonate derivatives with the following formula:—

These he prepared by partial sulphonation of the nitrite in known ways. They both yielded crystals of a hydroximidosulphate when dissolved in a little water and differed in no essential particular from nitrito-hydroximidosulphates. From hot solutions of nitrite and a hydroximidosulphate we obtained by cooling an apparently homogeneous crop of crystals of almost the same composition and properties as one or other of Raschig's salts. Raschig gave two ways for preparing the salt having the second of the formulæ just given and in these ways we have obtained the nitrito-2/3 normal hydroximidosulphate already described in this paper, but mixed with a little potassium sulphite. This impurity accounts for the alkaline reaction of Raschig's preparation and the presence in it of a little more than K<sub>3</sub> to S<sub>2</sub>.

He got the other salt (K<sub>2</sub> to S) only once and in the form of white crusts when working unsuccessfully for hydroximidosulphate in Claus's way, the other main product being imidosulphate, that is hydrolysed nitrilosulphate as he himself pointed out. We have obtained—also by sulphonating nitrite, following Fremy—a product qualitatively like Raschig's salt though quantitatively a little different from it, and at the same time like the second salt compounded of nitrite and 5/6 normal hydroximidosulphate, described by us on page 218. The percentages found by Raschig were potassium, 36.84, and sulphur, 15.50.

When Raschig's salt was dissolved in water and acidified it gave nitrous oxide as the only gaseous product while ours gave also some nitric oxide. This fact might have served to render incorrect the application of our formula to his salt but for the evidence there is that this was mixed with a little sulphite which would have reduced any nitric oxide. Its mother-liquor on further evaporation gave, we are told, so much sulphite along

with the next crop of the salt itself as to cause its rejection. The presence of sulphite in less quantity in the first crop of crystals will have been masked by the oxidising action of the nitric oxide in becoming nitrous oxide. That sulphite was present in Raschig's preparation well accords also with the fact that potassium hydroxide added in excess precipitated potassium sulphite, for, although hydroximidosulphate is itself decomposed by the most concentrated solutions of potassium hydroxide into sulphite and nitrite, this decomposition is slow and the sulphite only deposits after some time. Raschig's preparation when dried on a tile was only a powder, that is, presumably, was not obviously crystalline, a point which also indicates an impure salt. Since the potassium and sulphur are in the same ratio in the two salts, quantitative analysis would hardly have made its presence known.\* Inspection of Raschig's formulæ is of itself sufficient to prevent their getting accepted as in accordance with the facts. For from these formulæ both salts should be strongly alkaline, while in reality one is neutral. Above all it is hardly credible that dissolution in cold water should suffice to cause monosulphonated nitrogen to become disulphonated.

Raschig held his two salts to be identical with Fremy's potassium sulphazite and sulphazite respectively; but the nature of Fremy's salts will be found, we believe, more precisely given in the paper following this. The point we would here insist upon is that Raschig's preparations, judged by their chemical behaviour, have no claim to be considered as dihydroxylamine derivatives, being in every way indistinguishable from synthe-

<sup>\*</sup>Of the 3KNO<sub>2</sub> of our formula (p. 218) only one mol. can give nitric oxide and only to the extent of two-thirds of its nitrogen; the other third becoming nitric acid. Raschig's analysis indicates the presence of only 3/4 mol. active nitrite. The quantity of hydrated sulphite required to be present is therefore only 5.2 per cent, of the mixed salts.

tically prepared compounds of nitrite and hydroximidosulphates. Dihydroxylamine salts have as yet only a hypothetical existence and are likely to remain so. For the double linking of the oxygen atom with the tervalent or quinquevalent nitrogen atom seems always experimentally to make or break itself in a single act, notwithstanding its bipartite character.

Raschig in his researches on Fremv's sulphazotised salts got, besides those we have just discussed, two other salts of undetermined constitution, both of which were most probably also nitrito-hydroximidosulphates. They may therefore be noticed here although Raschig did not represent them to be dihydroxylamine derivatives. Yet they were evidently closely like the other two in properties. One was isomeric with potassium hyponitrososulphate (Pelouze's salt) and also with his (K2 to S) 'dihydroxylamine' salt, allowing for different hydration, and the other was isomeric with potassium 5/6 normal hydroximidosulphate. Each could be obtained but once and they only call for any detailed notice because of the theoretical importance given to them as isomerides of other salts. The first referred to above was mistaken by Raschig for Pelouze's salt (hyponitrososulphate) but that salt it certainly was not (this Journal, 9, 85). It was got by dissolving nitric oxide in solution of potassium sulphite and hydroxide and evaporating to a small volume till crusts formed. If we assume that air or nitric peroxide was not excluded there were the conditions present for getting a nitritohydroximidosulphate, for, as we show in a paper which will shortly follow this, nitrous fumes passed into potassium sulphite

solution generate hydroximidosulphate freely together with nitrite.

The other salt isomeric with 5 6 normal hydroximidosulphate was obtained in Raschig's attempt to form 2/3 normal salt by passing surphur dioxide into a solution of potassium nitrite and hydroxide and letting stand for a day. These, too, are conditions for geting nitrito-hydroximidosulphate. Now, both products agreed in being decomposed by water in such a way as to yield hydroximidosulphate and in other ways behaved as compounds of nitrite with one of these salts. The behaviour of the one isomeric with hyponitrososulphate was indeed exceptional in that when dissolved in water containing a little alkali it gave the 2/3 normal hydroximidosulphate when according to our calculation it should have given the 5 6 normal salt, while it also gave in hot alkaline solution a little nitrous oxide which only hydroxyamidosulphate is known to give. These peculiarities we may attribute to partial hydrolysis having occurred in the very unstable salt before these experiments were made.

The calculated formula for the isomeride of hyponitrosulphate as a nitrite compound is  $3KNO_2$ ,  $K_5H$  ( $NS_2O_7$ )<sub>2</sub>,  $2OH_2$ , and such a compound we have described on page 218; that for the isomeride of the 5.6 normal hydroximidosulphate treated as being a nitrite compound is  $3KNO_2$ ,  $6K_2HNS_2O_7$ ,  $5K_5H(NS_2O_7)_2$ , which in water should give crystals of  $K_2HNS_2O_7$ ,  $2OH_2$ . This compound salt we have failed to get but its occurrence can be readily accepted as possible. Its assumed existence affords a much more satisfactory explanation of the nature of this salt of Raschig's than that we were able to offer in our paper on hydroximidosulphates already referred to.

Nitroso isomer, found,	Potssm. 35.72	Sulphur 14.40	per cent.
Calculated,	36.05	14.75	,,
D. & H's salt, found,	36.17	15.05	"
Oximido isomer, "	33,04	21.23	,,
Calculated,	32.91	21.54	,,

For the present, the existence of isomesides of Pelouze's salt and Fremy's basic sulphazotate must be regarded as no longer even probable.



# Identification and Constitution of Fremy's Sulphazotised Salts of Potassium, his Sulphazate, Sulphazite, etc.

Bv

Edward Divers, M. D., D. Sc., F. R. S., Emeritus Prof.,

and

Tamemasa Haga, D. Sc., F. C. S., Professor, Tōkyō Imperial University.

A sufficiently concentrated solution of potassium nitrite and hydroxide submitted to the action of sulphur dioxide gave Fremy minute silky needles of a salt which he provisionally named potassium sulphazate. With slightly diminished concentration of the solution he generally obtained instead the brilliant, often hard, rhombie prisms of potassium basic sulphazotate (5/6-normal hydroximidosulphate, this journal, 7, 15). But sometimes there was obtained neither of these salts before the solution became transformed into a starch-like jelly through the formation of a salt which he named potassium metasulphazate, or else became filled with spangles of yet another salt called by him potassium metasulphazotate. When the solution was a little too dilute to give any of these and when too much alkali had not been added, there usually appeared peculiarly pointed crystals of the salt he named potassium neutral sulphazotate (2/3-normal hydroximidosulphate Rasehig) and, lastly, with still greater dilution

the minute brilliant needles of his potassium sulphammonate (nitrilosulphate Berglund). Still other salts he believed to be produced in the first stages of the reaction between the nitrite and sulphur dioxide, one of which he named potassium sulphazite; but this he did not obtain directly, finding a reason for this in the exceeding solubility of this early formed salt. He prepared it—but only in quite small quantity and as crystalline warty granules—by the action of water upon the 'sulphazate' whereby this was converted into 'basic sulphazotate' which deposited and a solution that on evaporation yielded the 'sulphazite.' These two salts could together in solution be changed back into the 'metasulphazotate' while the 'sulphazite' and the 'sulphazate' could similarly often be changed into the 'metasulphazate 'again. These two 'meta' salts he regarded therefore as perhaps merely double salts of the others. The 'sulphazite,' the 'sulphazate,' and the 'sulphazotates' he treated as being members of a series of salts in which there were to two atoms of nitrogen from one up to eight atoms of sulphur,—three in the "sulphazite, four in the 'sulphazate', and five in the 'sulphazotates.' With this conception of the nature of these salts, based on his analyses, it was easy to understand the decomposition of the 'sulphazate' into the 'sulphazite' and the 'sulphazotate.' But this and other of Fremy's interpretations of the facts observed by him have lost all importance and particular interest through the progress of chemistry since his memoir was published and only his account of the facts requires consideration now.

Subsequent work by others and ourselves in the same field has shown that Fremy in the account he gave of the preparation of his many salts went two little into details as to the conditions under which they were obtained,—apparently because he was not able to be more precise. When Claus attempted to get Fremy's salts he obtained only masses of minute crystals of salts of whose individuality and nature he could make out little because of the impossibility of dissolving them all up undecomposed. In his experiments the 'sulphammonate' (nitrilosulphate) was always formed in considerable quantity either as a first or secondary product and by its presence prevented any satisfactory investigation of the other salts. In Fremy's working, this most easily formed salt came only as the final product of the sulphonation and therefore gave him no trouble. Claus emphatically displayed his scepticism as to Fremv's results; vet in nearly every point in which he differed from Fremy as to the facts we find Fremy When Raschig repeated Fremy's work to have been right. but with the modifications in procedure introduced by Claus—he got results similar to, though less unsatisfactory than, those Claus had obtained. He made an approach to Fremy's work in so far as that he often got very little nitrilosulphate; nevertheless he too failed in his attempts to prepare the 'sulphazate' in Fremy's way.

In perhaps all essential points we can lay down the method to repeat Fremy's experimental work successfully. But in some cases a little uncertainty obtains owing to the fact that the very concentrated and complex solutions which yield Fremy's salts are apt to deposit what is virtually the same salt in different forms as well as at times salts quite distinct from each other under only slight and obscure variations in the circumstances attending their formation.

Sulphazate.—This is Fremy's first salt directly obtained in his sulphonation of the nitrite. In getting it he took approximately 5 mols. potassium nitrite to 2 mols. potassium hydroxide and a little water and into the solution passed sulphur dioxide

until it became almost filled with silky needles very soluble in water. So far it is easy to follow Fremy with a full measure of success if only the water used is limited to perhaps twice the weight of the nitrite and that the heating effect of the nitrite is counteracted by cooling. Claus and after him Raschig failed but then inexplicably to us they did not start with Fremy's proportions of nitrite to hydroxide, though even with the proportions they took, success was possible with care. The salt thus formed by Fremy was not tested and analysed by him until after it had been changed (but without his having recognised the fact) by the further treatment to which he submitted it. Before its change it is potassium nitrito-2/3 normal hydroximidosulphate described in the preceding paper, a neutral salt decomposed by water into its constituent salts. Fremy's finished 'sulphazate' was strongly alkaline and very caustic and when decomposed by water gave nitrite and the 5/6 normal hydroximidosulphate not the 2/3-normal salt. Also the analysis he gave of it furnished numbers such as the original product could not have given him. Instead of potassium, 33.10, sulphur, 18.06, and nitrogen, 7.9 per cent., he got potassium, 34.90, sulphur, 19.55, and nitrogen, 4.9. We can learn what his after-treatment was by reference to other parts of his paper where he speaks of the care necessary (when sulphonating the nitrite) to maintain the alkalinity of the solution by adding potassium hydroxide from time to time and of dissolving sulphazotised salts for examination in water containing this alkali. Certain it is he must have added some potassium hydroxide to the solution after getting it to crystallise, as a precaution to preserve the salt. Now the effect of this addition is to change the composition of the product without much affecting its silky asbestus-like appearance. The change in composition is to deprive it of much of its nitrite and to convert the 2/3-normal into more nearly normal hydroximidosulphate—to replace, therefore, potassium nitrite by potassium hydroxide. Accepting Fremy's mean numbers as accurate, what he analysed had the composition,

 $11K_3NS_2O_7,OH_2;K_2HNS_2O_7,2OH_2;2(K_2HNS_2O_7,KNO_2).$ 

	Potssm.	$\operatorname{Sulphur}$	Nitrogen	Alk. petssm.
Found,	34.9	19.55	4.9	— per cent.
Calc,	34.9	19.51	4.9	9.36 .,

But his analyses have no claim to receive such close treatment, his nitrogen seemingly being always much too low; and it is sufficient to say of his 'sulphazate' that it was the silky asbestus-like nitrito-2/3 normal hydroximidosulphate more or less converted into the also silky asbestus-like normal hydroximidosulphate, an account of it with which Fremy's description of its other properties entirely agrees. With dilute acids it gave slowly nitrous oxide unmixed with nitric oxide. Fremy specially points out that no sulphazic acid or any other sulphazates could be obtained from the potassium salt. There is, therefore, nothing to justify belief in this compound being the salt of a particular single acid, the sulphazic.

Sulphazite.—What Fremy named potassium sulphazite he only once obtained, and then not by direct sulphonation of the nitrite, in the form of white mammillated crystalline crusts from a solution thickened by the other salts contained in it. That is, to say, his sulphazate when dissolved in a little water containing some potassium hydroxide deposited crystals of basic sulphazotate (5'6 normal hydroximidosulphate), and left a mother-liquor which on cold evaporation till syrupy yielded the sulphazite. It showed great analogy with his sulphazate but was distinguished from it

by having little tendency to hydrolyse and by at once evolving some nitric oxide when its solution was mixed with a dilute acid. Water decomposed the *sulphazite*, but into what products was not ascertained.

We have sufficiently realised Fremy's expectations that his sulphazite might directly result from sulphonating the nitrite with subsequent addition of alkali. The substance obtained in this way did not differ greatly in composition from his:

	Potassm.	Sulphur
Fremy's salt,	38.16	16.27 per cent.
D. & H's salt,	36.94	16.37 ,,

and agreed with it in chemical properties, so far as is known. At the same time it was indistinguishable from a compound of nitrite with 5/6 normal hydroximidosulphate, and has been described by us as such in the preceding paper (p. 218) in which it stands as the third of these double salts and in which its preparation is given. Other experiments of various kinds have yielded us such 'mammillated crusts' as Fremy got, which, though only in rough agreement in percentage composition with his sulphazite, behaved like it and proved to be impure double salts of nitrite with 5/6 normal or more nearly normal hydroximidosulphate. We are therefore convinced that his sulphazite was only such a double salt.

Metasulphazate.\*—In Fremy's experience it sometimes happened, when passing sulphur dioxide into solution of nitrite and alkali of a concentration intermediate to that giving sulphazate and that giving basic sulphazate, the solution set to a starch-like jelly instead of crystallising. He obtained a similar jelly by cooling

<sup>\*</sup>Often, misprinted metasulphazotate in the French original, but not in the German translation.

a concentrated solution of *sulphazate* and *sulphazite*; also by boiling a solution of *sulphazate* and then cooling it. When strongly compressed the jelly became a transparent wax-like mass. Heated in this waxy state to 50°-60° it suddenly changed into a solution of *sulphazite* and minute crystals of *basic sulphazotate*. In all other respects it proved to be intermediate in properties to *sulphazate* and *sulphazite*. No other *metasulphazates* could be prepared from it, so that Fremy was disposed to regard it as being a double salt of *sulphazate* and *sulphazite*. Its constitution must therefore have been that of nitrite combined with normal or 5/6 normal hydroximidosulphate in such proportions and with such additions perhaps of alkali as prevented crystallisation.

We have not had Fremy's success in getting this salt in form of jelly and wax but have met with just such phenomena when forming barium sodium hydroximidosulphate, BaNaNS<sub>2</sub>O<sub>7</sub>, OH<sub>2</sub>, as will be found described in our paper already frequently referred to. We have however obtained a salt, or homogeneous mixture of salts, of the same composition as the metasulphazate, but with the form of the silky radiating fibrous crystals of the nitrito-normal hydroximidosulphate, from which it differed only in showing deficiency of nitrite, that is, it was equivalent in composition to a mixture of the normal salt and its nitrite compound, both of which crystallise with the same habit. We give below Fremy's numbers, our own, and those calculated for the expression, 3(KNO<sub>2</sub>, 2K<sub>3</sub>NS<sub>2</sub>O<sub>7</sub>, 4OH<sub>2</sub>); K<sub>3</sub>NS<sub>2</sub>O<sub>7</sub>; 3OH<sub>2</sub>.

	Potssm.	Sulphur	Nitrogen	Alk. potssm.
Found (Fremy),	35.10	16.74	4.81	—— per cent,
,, (D. & H.),	35.10	16.68		10.47 ,.
Calculated,	35.06	16.74	5,23	10.23 ,,

We got the salt by dissolving the hydroximidosulphate in

hot concentrated nitrite solution containing alkali. To 100 ec. water there were present  $45^{1}/_{2}$  grm. nitrite and  $1^{2}/_{3}$  grm. potassium hydroxide; for 66 mol. nitrite there were dissolved 10 mol. anhydrous normal hydroximidosulphate. But for the salt being in beautiful asbestus-like fibres, there was nothing to distinguish it from the jelly and the wax-like metasulphazate, which, therefore, we do not hesitate to class as a nitrito-hydroximidosulphate.

Basic sulphazotate, which Fremy considers next, has been shown by us already (loc. cit.) to be the 5/6 normal hydroximidosulphate, and not the salt of a distinct acid, the sulphazotic. It is liable to contain a small excess of potassium when crystallised from a strongly alkaline solution. A solution of the normal salt readily deposits it, as does also that of the nitrite compound of the normal salt.

Neutral sulphazotate was shown by Raschig to be the 2/3 normal hydroximidosulphate. The potassium sulphazotates were distinguished by Fremy from the salts previously described by him by their ability to form other sulphazotates by double decomposition. Fremy's analytical results in the case of the neutral sulphazotates are hopelessly out of accord with its constitution and properties, though those for the basic sulphazotate are satisfactory enough.

Sulphazidate, produced by the hydrolysis of the sulphazotate, is hydroxyamidosulphate (Claus). Sulphazilate and metasulphazilate, oxidation products of sulphazotate are ON(SO<sub>3</sub>K)<sub>2</sub> and ON(SO<sub>3</sub>K)<sub>3</sub>, and have been studied by Claus, Raschig, and Hantzsch.

Metasulphazotate.—Sometimes Fremy got a salt in the form of spangles (paillettes), in appearance like minute crystals of basic sulphazotate, but differing from these in not being hard under pressure. This salt he named, therefore, metasulphazotate. According

to him it is also obtainable by mixing (hot) solutions of the (basic) sulphazotate and sulphazite. It is very soluble in water, very alkaline and unstable unless the water contains alkali. In pure water it becomes basic sulphazotate and sulphazite again. shows the greatest analogy with metasulpharate and is distinguished in the same way as this salt from basic sulphazotate. may be a compound of basic sulphazotate and sulphazite. So far Fremy. It will be evident that there is nothing in its history or properties to distinguish it, except its occurring in the form of sparkling particles and even that can be met with in the basic sulphazotate suddenly precipitated; we have also got other of the sulphazotised salts in what may be called spangles, though not this particular salt. In the preceding paper, page 214, we have described an impure form of nitrito-2/3 normal hydroximidosulphate obtained by dissolving the 5,6 normal salt in a hot concentrated solution of nitrite, but still not so very concentrated as to give the nitrito-56 normal double salt. This preparation is lustreless while in its mother-liquor, but when dried on the tile has a fine silvery lustre. It has when dried in the desiccator exactly the composition of Fremy's metasulphazotate and is much less alkaline than the metasulphazotate and is much less alkaline than the metasulphazate. It may be formulated as

 $K_3NS_2O_7$ ;  $9(KNO_2, K_2HNS_2O_7, 1^{1/2}OH_2)$ .

	Potssm.	$\operatorname{Sulphur}$	Nitrogen	Alk. potssm.
Found (Fremy),	33.8	18.6	3.5	—— per cent.
,, (D. & H.),	33.79	18.35	<del></del>	1.09 ,.
Calculated,	33.68	18.37	7.63	1.12 ,,

Sulphammonate and sulphamidate are respectively nitrilosulphate and imidosulphate (Berglund).





# On a Specimen of a Gigantic Hydroid, Branchiocerianthus imperator (ALLMAN), found in the Sagami Sea.

Bv

**M. Miyajima**, *Rigakushi*. Science College, Imperial University, Tōkvō.

With Plates XIV & XV.

On the morning of January 1, 1899, quite a commotion was produced in the Marine Biological Station at Misaki by the bringing in of a very beautiful and gigantic Coelenterate (Pl. XIV). It had been caught, on the previous day, by a fishing "long-line," from a depth of about 250 fathoms near Okinose, a submarine bank 18 kilometers south of Misaki. It was an object which was calculated to raise enthusiasm in a naturalist. A large disc surmounted a long stalk which evidently fixed the animal on the sea-bottom. A circle of numerous graceful tentacles hang down from the margin of the disc, while on its upper surface arose an oral tube, surrounded at its base by bushy dendritic appendages and having a second circle of slender tentacles around its upper edge. The total height of the animal was 700 milli-

meters and the prevailing colour transparent scarlet. It was agreed on all sides that it was a New Year's gift from Отоніме\* and that it should be known in Japanese as Otohime no Hanagasa.

The specimen, when brought in, was entirely fresh but was not living. It was placed in 2% formalin to preserve, if possible, something of its beautiful colour. At first the attempt seemed successful, but after a while the colour began to fade gradually, until now the specimen is completely bleached to pale white. For histological examination, pieces of the tentacles and the dendritic appendages were fixed in the sublimate and in Perenyi's fluid.

The specimen was handed over by Prof. MITSUKURI to me to work out its finer structure.

It was evident from the first that the specimen was very similar to the form only a short time before described by MARK ('98) as *Branchiocerianthus urceolus*. I started, therefore, with an idea that I was dealing with an Actinian.

As I proceeded in my investigation, however, it became plain that this idea was not tenable, and the conclusion was finally reached that the animal was very closely allied to Corymorpha, and that it belongs probably to the species obtained by the "Challenger" at about the same locality and named by Allman ('85) Monocaulus imperator, notwithstanding many discrepancies between his description and the specimen. This conclusion was communicated through Prof. Mitsukuri to Dr. Mark and a request was also sent to him, that during his opportune stay in Europe, he should,

<sup>&</sup>quot;Otohime" is a beautiful goddess who is supposed to have her palaces at the bottom of the sea. "Hanagasa" is the flower-sun-shade or ornamental parasol. Thus Otokime no Hanagasa means "the ornamental parasol of Otohime."

if possible, examine the original specimen of *Monocaulus imperator* in the British Museum. To the results of his examination of the specimens I shall return in the later part of this paper.

Meanwhile an article was published in the Zoologischer Anzeiger by O. Carlgren ('99) throwing doubt on Mark's Branchiocerianthus being an Actinian, and contending that it more probably is a Corymorpha or at least a form standing very close to Corymorpha.

In June, 1899, a correction was published by MARK ('99) himself in the Zoologischer Anzeiger. His previous preliminary description had been based on external anatomy, and he now frankly admitted that further researches had convinced him of the fact that the animal in question must be more nearly related to the Hydroidea than to the Actinia, though its exact affinities he had not yet determined. In a postcript he mentions our conclusions which had been communicated to him, as mentioned, by letter, and thinks that both his and our specimens belong to the same genus and that our specimen is probably identical with the Monocaulus imperator of Allman.

Before going further I wish to express my deepest feeling of obligation to Prof. MITSUKURI for the supervision and advice which he has given during the progress of my work.

## Description.

This hydroid is a solitary form consisting of a well marked hydranth and a hydrocaulus. Its most striking feature is a strongly expressed bilateral symmetry. The hydranth is discshaped and bears two sets of tentacles and a circle of dendritic gonosomes, all showing in their arrangement a well marked bilaterality. The hydrocaulus, which is attached not to the center but to the edge of the hydranth, is nearly cylindrical and increases in diameter from the attachment of the hydranth towards the end which is fixed in the sandy sea-bottom. The total height of the animal attains 700 mm., as measured from the top of the oral tube to the attached base of the hydrocaulus.

In the fresh condition the hydranth was rose pink and its tentacles, both oral and marginal, were deep scarlet in colour, while the gonosomes possessed light rufous colour. The hydrocaulus was light pink in colour, being quite pale in its middle.

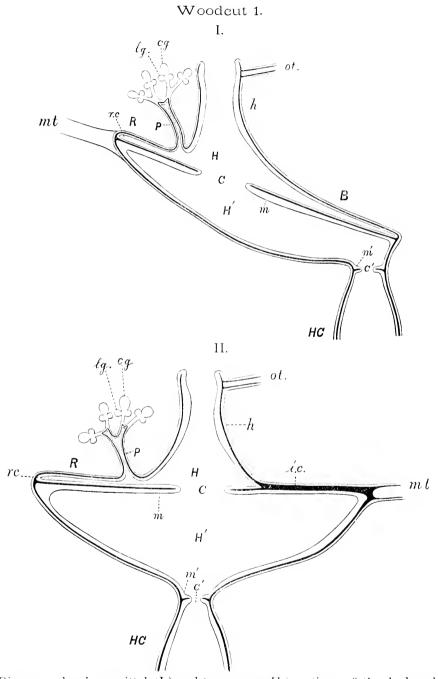
The general features and the colours are well shown in Fig. 1, Pl. XIV, which was drawn from the preserved specimen by Mr. NAGASAWA, artist of our Institute, making use also of the rough sketches I made at the time of the fresh object.

### Hydranth.

The upper surface of the hydranth is flattened so that it may be described as an "oral disc." The lower surface, however, assumes a shallow funnel-shape, which passes downwards into the hydrocaulus. This disc has an oval outline, but differs from that of *Branchiocerianthus urceolus*, in having its sagittal diameter less than its transverse, the two diameters being respectively 80 and 90 mm. (Woodcut 2).

At one end of the sagittal diameter is attached the hydrocaulus where the circle of the marginal tentacles is also interrupted. The plane of the disc is oblique to the long axis of the hydrocaulus (Woodcut 1, I), though not to the same degree as in *Branchiocerianthus urccolus* Mark.

The edge where the hydrocaulus is attached I shall designate the lower, and the opposite the higher, edge.



Diagrams showing sagittal (I.) and transverse (II.) sections of the hydranth. B. hypostomal region of the disc; C orifice of the diaphragm (m) in the hydranth; C orifice of the diaphragm (m') in the hydrocaulus; eg. central, lg. lateral, globule of the gonophore; H. npper, H lower cavity of the hydranth: h. hypostome; ic. intercalated cord; HC hydrocaulus; mt. marginal, ot. oral, tentacle; P. peduncle of the gonosome; R. outer region of the disc, provided with the radial canals (c.c.).

The hypostome (Woodcut 1, I, II, h), the superior prolongation of the disc, is slightly conical, diminishing gradually in its diameter from the base towards the free end where the mouth opens. A little below the mouth the hypostome bears a brush-like group of about 180 filiform tentacles (ot.) which are arranged in three or more closely packed verticils, the outer tentacles being much larger than the inner. The outermost ones attain a length of 50-55 mm., while the innermost are so small and crowded that I could neither measure them well nor count their exact number. Below the oral tentacles the hypostome is slightly constricted, but there is no indication of syphonoglyph which is said to be present in the oral tube of Branchiocerianthus urceolus. The side of the hypostome turned towards the lower edge of the disc passes gradually to the disc, while on the opposite side it seems abruptly raised from the disc, so as to make an angle between. The hypostome is thus oblique to the disc proper which again is not perpendicular to the axis of the hydrocaulus. Hence we can show the relation of the three parts, the hypostome, the disc and the hydrocaulus, diagramatically with three lines, of which two vertical ones, corresponding to the axes of the hypostome and of the hydrocaulus, meet with an oblique one representing the axis of the disc, forming obtuse angles between them (Woodcut 1, 1).

The base of the hypostome (Woodcut 2, B.) occupies about the middle of the disc, but on the side turned towards the lower edge, its base gradually becoming lower and lower, may be said to stretch as far as the margin of the disc, while laterally and towards the higher edge it is distant from the margin 35 mm. and 22 mm. respectively. It thus assumes an ovoidal outline, the pointed end attaining the lower margin of the disc and passing directly to the

#### Woodcut 2.

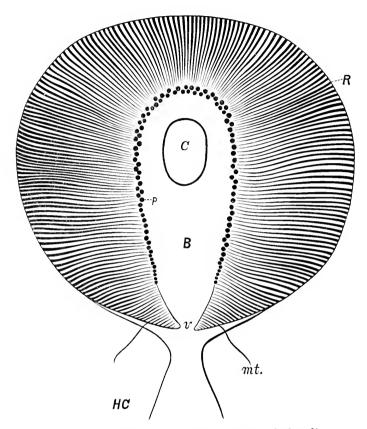


Diagram showing the upper surface of the disc. v, hiatus at the lower edge of the disc. Other letters as in Woodcut 1.

surface of the hydrocaulus. The longer (i. e. up—down) diameter of this ovoidal space measures 60 mm., while the transverse at the widest middle portion is only 25 mm. This space is destitute of the radial canals which are prominently seen in the remaining part of the disc.

Around the margin of this hypostomal region there arises from the surface of the disc a row of dendritic gonosomes (p) which in shape strongly remind one of the heads of cauliflowers.

They number in all 96 and are arranged approximately in a single row, which, being interrupted at the lower edge of the disc, assumes the form of a horse-shoe. (Woodcut 2, P). At the two ends of the horse-shoe are situated the smallest gonosomes which stand at a distance of 15 mm. across from each other. The length of the stalk of the gonosomes varies from 20 mm. to 60 mm. While the gonosomes nearer the lower edge of the disc are on the whole shorter than those nearer the upper edge, it is to be noticed that the larger and smaller gonosomes are placed alternately, indicating faintly the two circles in their arrangement, the larger gonosomes being placed in the outer, and the smaller in the inner, circle.

The region of the disc outside the gonosomes is marked with numerous radial canals (Woodcut 2, R.) which run from the base of the gonosomes to the margin of the disc. This region thus assumes the form of a wide horse-shoe, whose two arms gradually diminish in their breadth towards the lower edge of the disc until they terminate at that edge. Hence this region varies in breadth, measuring 20 mm. on the median line at the higher edge, and 35 mm. on the lateral region, while on the lower side both arms are practically zero.

The radial canals (Pl. XV, Fig. 1, r.c.) slightly swell out the surface of the disc thus giving the latter an undulating appearance. The canals are intercalated by solid cords (Pl. XV, Fig. 1, i.c.) which appear on the surface of the disc as opaque lines. The canals and the intercalated cords are longest in the lateral region where they run obliquely across the disc, and are longer than the breadth of this region. The canals situated nearer the lower edge are smaller and shorter than those higher up, until at the both arm-ends they are practically nil. On the other hand the

canals on the higher side of the disc are not so long as those on the lateral, but run straight from the base of the gonosomes to the outer margin of the disc, the length of the canal being thus the same as the breadth of this region (Woodcut 2).

The radial canals and the intercalated cords increase in their width towards the outer margin of the disc where the both structures are broadest. Inwards, the radial canals open into that part of the hydranth-cavity where the cavities of the gonosomes stand in communication with the latter. Outwards, the canals terminate blindly on the margin of the disc. The intercalated cords enlarge suddenly near the margin of the disc and acquire a cavity which forms a part of that of the marginal tentacle (Pl. XV, Fig. 1).

The name of marginal tentacles (mt) is given to the outermost circle of filiform tentacles arranged like a fringe around the margin of the disc. The circle is not complete, there being a hiatus (v) at the lower edge of the disc where the surface of the hypostome passes directly into that of the hydrocaulus. shortest tentacles arising from the 6th or 7th intercalated cord, counting from the lower edge, occupy the two ends of this incomplete ring. Whether there were any smaller tentacles nearer the lower edge, I am not sure. There is no indication, so far as I can see, of any having existed. Towards the higher edge of the disc they increase successively in length until about the 10th (counting from the lower edge) is reached, of which the length on both sides is about 200 mm. After this there seems to be no special arrangement of the tentacles, which vary from 200 mm. to 300 mm. in length. They numbered 198 in all. The tentacles are flattened at their base, and are compressed so closely with one another that the basal portion appears to form

a part of the disc. Just above the flattened base the tentacle assumes the form of a tube, 4 mm. in diameter, and tapers gradually towards its free end.

The hydranth (Woodcut 1, I. II.) contains a wide cavity which is separated by a thin membrane (m) into two parts, an upper (H) and a lower (H'). The superior prolongation of the upper cavity is that of the hypostome, which does not show any indication of the septal partition. The lower cavity is more spacious than the upper, and not only occupies the whole lower part of the hydranth but extends also through the entire length of the hydrocaulus.

The membrane (m) separating the hydranth-cavity arises diaphragm-like just below the upper wall of the disc. In about the center of this diaphragm, directly below the mouth, is an ovoid orifice (Woodcut 1, II, C) which puts the upper and lower cavities in communication with each other. The orifice is 11 mm. and 15 mm. respectively in its transverse and sagittal axes.

That part of this diaphragm which corresponds to the part of the upper surface of the disc marked B in Woodcut 2, i.e. to the basal part of the hypostome, projects into the eavity of the hydranth like a shelf, with the aforesaid opening near its middle and with no attachment either above or below. Outside this portion, however, the diaphragm forms the floor of the radial canals mentioned above, so that it is suspended, so to speak, by the numerous intercalated cords (vide supra) to the upper surface of the disc. At the margin the diaphragm is united to the outer wall of the hydranth (Woodcut 1).

To show the somewhat complicated relations existing between the marginal tentacles, radial canals, intercalated cords, etc., a series of sections (Pl. XV Figs. 4-9) passing through the lines 1-1, 2-2, 3-3, 4-4, 5-5, 6-6 in Fig. 1, Pl. XV is introduced. The first section (Fig. 4) through the outermost margin of the disc, which corresponds to the line 1-1 in Fig. 1, shows that the bases of the marginal tentacles (t.b.) and the blind ends of the radial canals (r.c.) are arranged alternately, the former projecting out above and below more than the latter. The upper projection corresponds to the enlarged end of the solid cord. The cavity of the tentacle is almost filled up by the spongy endoderm which lines the whole cavity of the animal, so that it remains as a narrow canal only in the upper and lower swollen parts of the tentacles. On the other hand the radial canals contain a wide cavity which is clearly separated from that of the tentacle-base by the well developed mesoderm. In the next section (Fig. 5, through the line 2-2, Fig. 1) cut just inside the margin of the disc, the radial canals already assume their characteristic shape in crosssection, while the intercalated cords have already lost their cavity entirely. Bounded by the mesoderm the intercalated cord assumes in cross-section the form of a trapezoid. It is convenient to distinguish here three kinds of the mesoderm-lamellæ, the upper, basal, and the vertical. The upper lamella (u.l.) is situated along the surface of the disc, the basal (b.l.), in the floor (i.e. in the diaphragm), and the vertical (v.l.) connects these two lamellae. When traced inwards, (Pl. XV Figs. 6, 7) the intercalated cord becomes thinner and thinner, until it no longer shows in crosssection the form of a trapezoid, but assumes the shape of a triangle formed by two vertical and one basal lamella. Where the gonosome arises (Fig. 8), the vertical lamella does not reach the upper lamella; hence the radial canals communicate here with one another and form the upper common cavity of the hydranth. Within the circle of the gonosomes (Fig. 9) the upper lamella stands entirely separated from the basal, on which the vertical lamella shows itself only as a ridge-like line which in cross-section is recognizable as a simple small knob.

Preserved in formalin, the fine tissues of the animal were unfortunately mostly gone. Luckily, however, the pieces of the gonosomes and the marginal tentacles, which were preserved in sublimate, etc., helped us to ascertain something of the histological character of the animal.

The wall of the animal-body, I need hardly say, consists of the three layers, ecto-, endo- and mesoderm as in other Coelenterates.

The ectoderm, the outermost layer, has been entirely shed off from the specimen in formalin, but in the pieces fixed with sublimate was well preserved. This tissue is a single layer of cylindrical cells which in their preserved condition are more or less vacuolised. There are present a few nematocysts which are characteristic of the ectoderm of Coelenterata.

The mesoderm is a very firm, supporting layer which is placed between the ecto- and endo-derm or two portions of the endoderm. This tissue was well enough preserved even in formalin so that the structure of the animal could be largely made out by this layer alone.

The endoderm, the innermost layer, which lines the whole cavity of the animal, remained unfortunately only here and there in the specimen in formalin. From these patches it could be made out that the endoderm lining the hydranth-cavity is several cells thick (Figs. 3 & 10). The cells are irregularly formed and contain but a little cytoplasm which is pressed towards the wall with the nucleus. Consequently the wall of the cavity gives a

spongy appearance. I can not think that this appearance of the endoderm is eaused by bad preservation, for the tentacles fixed with sublimate show also the same structure. In the preserved state, the endoderm forming the upper ceiling of the lower cavity of the hydranth has a thickness of 3 mm.

In the cross and longitudinal sections (Figs. 11 & 12) of the marginal tentacle, the whole of the space inside the mesoderm is entirely filled up with a tissue which reminds one of the vertebrate notochord. It has the same structure as the spongy endoderm of the hydranth-cavity already mentioned. Only near the base of the tentacle, this spongy tissue leaves in the center a small cavity which is separated by the mesoderm from the hydranth-cavity. Hence the cavity of the tentacle-base is of a limited extent, extending not farther towards the distal end, and communicating nowhere with the general cavity of the hydranth. A longitudinal section (Fig. 2) through the margin of the disc shows plainly the relations of the disc and the base of the tentacle (the mesoderm being drawn darker than other parts in the figures).

The gonosomes (Fig. 1, p.) as already mentioned consist of the branched tubular stalks, upon which the gonophores are grouped in a crowded cluster. Each stalk branches dichotomously into about the 10th or 12th order. Each branchlet terminates in a group of small globules, of which we recognize two kinds (Figs. 13 & 14). The one kind of which there is only one in each cluster is situated on the top of the terminal branch, while the others take a more lateral position. The former is larger than the latter, consisting of the irregularly shaped cells mostly vacuolised (Fig. 14, c.g.). In this kind of globule the mesoderm of the branch is no longer recognisable and the ecto-and endo-derm can not here be clearly distinguished. It seems,

however, reasonable to suppose that the centrally placed smaller cells which are continuous with the endoderm of the branchlet belong to that layer. The cells which presumably belong to the ectoderm and form the main part of this globule seem to be mostly distended. In this globule the nematocysts (Fig. 15, n.) are found in a large number; hence the central globule may be regarded as the battery.

The lateral globules (Fig. 14, l.q.) are mostly spherical and consist of compactly packed cells rich in cytoplasm. The mesoderm prolonged from the branchlet distinctly separates the ectoderm from the central cell-mass. After examining many sections I was able to find a few globules which enable us to see that the clusters are true gonophores. In such globules (Fig. 16), one is able to see that the ectoderm cells at the tip are grouped into a mass forming the "bell-nucleus" which pushes the endoderm in as a cup. This part of the endoderm is arranged into a regular layer one cell deep and is easily distinguishable from the remaining part. Owing to the section (Fig. 16) having been cut slightly obliquely, the cavity in the endoderm seems irregular and very limited. In reality, there is a wide cavity occupying the whole interior of the globule, which communicates with that of the branchlet. I could not detect gonophores developed any further than this in our specimen. January is probably not the season in which the ripening of the sexual products takes place.

The terminal branch thus bears two sorts of globules, the one being a nematocyst-battery and the other a true sexual organ. Hence the dendritic gonosome of this animal is a peculiar organ which bears on a common stalk the sexual and defensive elements.

In other hydroids these two elements are borne on separate stalks, as for example in *Pennalia*.

#### Hydrocaulus.

The under part of the hydranth is prolonged to a shallow funnel whose neck corresponds to the hydrocaulus. At about the point where the hydranth joins the hydrocaulus, there is a circular constriction (Woodcuts 1 & 2). Here the diameter of the hydrocaulus is only 9 mm. and from this part down to the base it increases in its diameter. Within the constriction is a diaphragm (Woodcut 1, I, II, m') separating incompletely the cavity of the hydranth from that of the hydrocaulus. In other words the circular constriction is the surface expression of the insertion of the diaphragm. In the midst of this partition there is an opening (Woodcut 1, I, II, C') which puts the two eavities above and below in communication. It is about 4 mm. in diameter and is almost circular. The plane of the diaphragm is not visibly oblique to the long axis of the hydrocaulus. In the specimens of Monocaulus imperator in the British Museum, this diaphragm is, according to Dr. Mark, distinctly oblique and the central opening is much elongated.

The hydrocaulus is a hollow tube which has a total length of 650 mm, including the proximal end with hair-like appendages. The hydrocaulus, even when fresh, was collapsed and more or less longitudinally folded, so that the exact measurement of its diameter was almost impossible. Approximately, it was 15 mm, just below the constriction, 25 mm, at the middle, and 42 mm, at the terminal root.

The outer surface of the hydrocaulus is smooth. In the upper

half of it there are visible from outside 15-20 longitudinal wavy bands (Fig. 17). They stand about 2-3 mm. distant from one another and run down to about the middle part of the hydrocaulus where they become obscure. From the surface they look remarkably like the mesenterial filaments of an Anthozoon. These wavy bands anastomose here and there with one another and give to the hydrocaulus of our specimen an appearance much resembling that of Corymorpha. Though the bands are in the preserved state still visible, they were more conspicuous when fresh. These longitudinal bands show themselves in cross-section (Fig 18) as dense spots (x) in the mesoderm, which have a great affinity for any staining agents. From the bad state of preservation of the specimen, in which the ectodermal and endodermal cells were mostly lost, I could not ascertain whether the wavy bands were the endoderm canals, a structure peculiar to Corymorpha, or not. I think it, however, very probable that they existed, and gave rise to these band-like appearances. In the published accounts of Monocaulus imperator of Allman the endoderm canals were plainly described and figured.

The mesoderm is very well developed, especially in the hydrocaulus where it reaches a thickness of about 0.2-0.3 mm. This remarkable layer shows itself in the form of a fibrillated membrane, which, when macerated with caustic potash, is separated into two layers, the outer longitudinal (Fig. 19, l.l.) and the inner circular (Fig. 19, c.l.). The former is thicker and stains less with any coloring matter than the latter.

In our specimen there is no sudden bulb-like expansion at the lower end of the stalk, such as is described by Mark in Branchiocerianthus urceolus or by Allman in Monocaulus imperator. The lowest and broadest part of the hydrocaulus is enclosed for about 30 mm. from the base in a chitinous sheath which gives an anchorage to the Hydrozoon. With the exception of the upper edge the sheath (Fig. 20 s) bears in most parts very numerous hair-like processes (ap) of brown color, which are so entangled that many foreign bodies (e.g. Echinus spines, sand grains, dead shells) are wrapped up within them. The sheath and the root proper are united so closely that they are not to be separated from each other without tearing. In contrast to the pink-colored hydrocaulus the brown color of the sheath with its appendages is very conspicuous.

At the lowest end of the hydrocaulus the wall is very delicate and has an opening, the margin of which is destitute of the hair-like appendages.

Above the root the mesoderm possesses here and there irregular small depressions which are recognized by tolerable magnification from the surface as clear spots. These depressions are also present in the wall of the root enclosed in the sheath.

A portion of the root cut longitudinally (Fig. 21) shows that the sheath with its appendages is separate from the root proper, but has an organic connection with it. The hair-like appendage (ap.), which is seen to be a slender hollow process of the sheath, embraces in its interior the thread-like outgrowth (o.) of the wall, which perforates the mesoderm and is connected directly with the endoderm of the inner cavity of the hydrocaulus. Hence it seems to me that the above-mentional small depressions in the mesoderm are certainly the indications of the wart-like processes of the wall of the hydrocaulus as in Corymorpha.

#### Summary.

- 1. The most striking feature in our specimen is its strongly expressed bilateral symmetry as shown by the excentric attachment of the hydranth to the hydrocaulus and by an interruption of the circles of the gonosomes, radial canals, and marginal tentacles at the lower edge of the disc. Those who have read the above account will, I think, agree with me in thinking that this bilateral symmetry is due, not to the primitive state of the body-organization, but rather to its elaboration and specialization. We must therefore regard this remarkable case of bilateral symmetry in a hydriform person as very different from that expressed for instance in the planoblast of Corymorpha and Dicoryne, which is but temporary and occurs only at a certain period of development, or from the biradial symmetry as expressed in a few genera like Monobrachium and Lar by a reduction in the number of tentacles.
- 2. The hydranth-cavity is divided into two parts, of which the upper is in its outer part again divided into many radial canals visible even on the surface of the disc. That remarkable structure is not, however, peculiar to our specimen. For example, the hydranth-cavity of *Tubularia* is divided similarly into two parts by a peculiar ring-shaped formation\* observed by several authors. In *Tubularia* the lower cavity is narrower than the upper, so that the former forms a slender canal in the middle of the "Wulst." Gosta Grönberg ('98) described in the hydranth of *Tubularia indivisa* slender endoderm-canals which are the same in number to the proximal (marginal) tentacles and situated between every two tentacle-bases, running obliquely from the com-

<sup>\*</sup>O. Hamann ('82) described that formation as "aboral Wulst," G. Grönberz ('98) as "Mesoderm-wulst."

mon cavity outwards and downwards. These canals, though not visible from the surface, may be regarded as corresponding to the radial canals in our specimen, since they both arise from the upper cavity of the hydranth and are arranged alternately with the marginal tentacles.

- 3. The tentacles are filiform and arranged in two sets, oral (distal) and marginal (proximal), as is characteristic of the tentacles of *Tubularidæ*, *Corymorphidæ*, and *Monocaulidæ*. The cavity of the tentacle is mostly obliterated, being filled up with a cellular tissue—a condition very frequently met with in the tentacle of the Hydrozoa.
- 4. The dendritic appendage is a true gonosome which bears in its summit the sexual elements. Our specimen seems to be immature, hence it could not be decided whether the gonophore is a planoblast or a sporosac.
- 5. The hydrocaulus is marked with many wavy bands visible from the surface, and possesses a thin sheath with filamentous appendages at its lowest end.

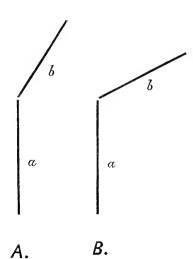
### Considerations on the Systematic Position of our Specimen.

Those who would compare the account given above of the structure of our specimen with that of Branchiocerianthus urceolus, Mark ('98) will not for a moment doubt that we have in these two cases essentially similarly constituted animals. It seems almost superfluous to call attention to the points of likeness: the hydrocaulus with the wavy bands in its upper half and with the sheath and filamentous appendages at its base, the hydranth surmounting the hydrocaulus, with its radial canals, dendritic gonosomes, and two sets of tentacles, all of which show a strongly expressed

bilateral symmetry, being interrupted at the lower (or Mark's posterior) edge where the hydrocaulus is attached.

That our specimen and Brachiocerianthus urceolus belong at least to the same genus, there can hardly be any doubt. Whether they belong to the same species is another question. It is perhaps premature to decide this point, at present, in as much as Mark has not yet published his full paper. Judging from his preliminary notice which deals exclusively with the external features, the following are the chief points of difference.

a. The general shape. B. urccolus is stated to have an extremely graceful, symmetrical vase-like figure with flaring lips. The lateral margins of the hydranth-disc were in the natural state "folded in symmetrically from either side, so as almost to touch at a point, a little below the middle of the oval. This bending in of the margins of the disk produces at the upper end of the animal a sort of eccentric funnel-shaped depression, " The fancied resemblance of



the animal to a little pitcher, which this side view presents has suggested the specific name adopted—urecolus." (MARK '98 p. 148). The pitcher or vase shape of the hydranth is thus due to two causes: (1) the folding in of the disc-margin, and (2) the extreme obliquity of the axis of the hydranth to the axis of the hydrocaulus. In the annexed woodcut, a represents the axis of the hydrocaulus and b

that of the hydranth. Thus these two axes make in *B. ur-ceolus* an extremely obtuse angle as in A, and thus help to produce the vase-shape. In our specimen the angle which these two axes make with each other (B) is much less obtuse, and moreover the folding in of the disc margin has not been noticed from the first, either in the fresh or preserved state. The disc lay flat and open as a disc, and never suggested the idea of a pitcher.

b. The shape of the disc is oval in B. urceolus; in our specimen it is more nearly circular. Moreover in the former, the sagittal or longitudinal diameter is greater than the transverse, while in our specimen it is the transverse diameter which is the greater of the two. The following measurements will make this point clear.

Sagittal (longit.) diam. in mm. $B.\ urccolvs.$	Trans, diam, in mm.	Ratio of trans. diam. to sagittal.
Small specimen25	15	60%
Large specimen38	30	nearly 79%
Specimen of Science College	90	112.5%

c. The size:—

Length of the hydrocaulus in mm.	Maximum length of the marginal ternacle in mm.	of the oral tentacle	
B. urccolus105-200	125	30-35	
Specimen of 650	300	50-55	

d. The lower end of the hydrocaulus:—Mark describes a bulb at the lower end of the hydrocaulus. In our specimen, there is no such sudden enlargement as deserves the name of a bulb, although that end is, as has been stated, the largest.

e. The radial canal:—Mark mentions that the radial canals of B. urceolus run "from the base of the oral tube to the bases of the marginal tentacles, before reaching which many of them fork, each of the branches communicating with the lumen of a single tentacle" (Mark '98, p. 150). The case is very different in our specimen in which the radial canals do not fork at all and do not communicate with the lumen of the marginal tentacles. The latter, on the contrary, are the continuations of the intercalated cords.

Whether these differences are to be regarded as only specific or due simply to the differences in size, age, etc., we must leave for the present an open question. I am inclined, however, to think that *B. urceolus* and our specimen are of different species.

References have already been made several times in the course of the foregoing pages to the resemblance of our specimen to *Monocaulus imperator* of Allman, a gigantic hydroid dredged by the Challenger off Yokohama (stat. 327). The description given by Allman of this animal in his report of the Hydroidea of that Expedition ('88) is not as exhaustive as is desirable. He makes no mention of any bilateral symmetry in the animal, but we must remember that the specimens which he had before him were extremely badly preserved, as he is careful to mention, and that the figure of the animal which was made on the spot by the artist of the Expendition must necessarily have been made hurriedly, and as we can testify from our own observation of the fresh object, it is very easy to overlook such a feature as bilateral symmetry when the disc is lying in the midst of a mass of tentacles. Of course the best thing we could do under the cir-

cumstances was to appeal to the original specimens. At the request of Prof. Mitsukuri, Prof. Mark, who was opportunely staying in Europe at the time, was kind enough to examine the type specimens of Monocaulus imperator, kept in the British Museum. The results of his observation were not entirely conclusive, as the specimens "have so long been in strong alcohol that it was quite impossible to make out anything very satisfactorily." He naturally made special efforts to ascertain the condition of the hydranth—whether it was radially or bilaterally symmetrical. In one specimen, he felt tolerably confident, though by no means sure, that there was an interruption narrower than in Branchiocerianthus urceolus in the marginal tentacles. In another specimen the central opening in the diaphragm which divides the cavity of the hydranth from that of the hydrocaulus was found much elongated—a point which in his opinion pointed to bilateral symmetry.\* He also thought that there is much less obliquity of the hypostomal region to the axis of the Hydroid than in B. urceolus "for the wall of the hydranth between the constriction and the base of the tentacles can be seen to be nearly the same height all around, or at least not markedly different on opposite sides." This last point is against the view that our specimen is identical with Monocaulus imperator, for although the disc is much less oblique in our specimen than in B. urceolus, as shown above, the hydrocaulus is attached at one end of the sagittal (longitudinal) diameter of the disc. But Prof. MARK adds, "the specimens were so much wrinkled and folded that I have not much confidence in this conclusion." There is

<sup>\*</sup> In our specimen the opening which puts the hydranth-cavity and that of the hydrocaulus in communication is not elongated, but almost circular as already stated.

one curious point of difference between our specimen and *Monocaulus imperator*. While the hydranth in the Challenger specimen is much smaller than that of our specimen, the stalk is enormously longer, being said to reach the almost incredible length of 7 feet 4 inches. This is, however, stated to be when stretched, and is not the normal length.

While it is not thus possible to establish absolutely the identity of our specimen with Monocaulus imperator of Allman, there are on the whole strong probabilities in favor of this assumption. Those who read carefully Allman's description will notice that the points which he brings out distinctly in the structure of his species, such as a wide cavity extending through the entire length of the stalk, the presence of the stalk-mesoderm in the shape of a fibrillated membrane—a point which Allman emphasizes as "the most striking feature in the histology of the Hydroid"—and so forth, are absolutely similar in our specimen. If we remember in addition that both came from practically the same locality, it is, I believe, within the scope of reasonableness to conclude that our specimen belongs to Monocaulus imperator of Allmann.

If this is really the case, we must examine other species in Monocaulus. The genus includes, besides Monocaulus imperator, two other species; M. glacialis, (Sars) (for which Allman established the genus) and M. pendula, (Agassiz). These two forms show, however, a radial symmetry, and now that M. imperator is shown to have a bilateral symmetry, can not possibly be put in the same genus with the latter. M. imperator must therefore be separated from the other two species and placed in a new genus. According to the rules of nomenclature, this new genus

must take the name Branchiocerianthus in first given by Mark, and our specimen then ought to be known as

Branchiocerianthus imperator (Allman).

P.S. We shall await with interest the full report on the specimen of *Monocaulus* obtained by Prof. Chun in his recent deep-sea expedition ('99).

December, 1899. Zool. Institute, Science College, Imperial University, Tokyo.



<sup>\*</sup>Mark ('99) mentions that Rhizonema carnea of S. F. Clarke ('76) may be a form same as, or closely related to, Branchioceriandus. Clarke's original description is very brief and it is impossible to determine whether Mark's suspicion is correct or not. At any rate, Clarke makes no mention of any bilateral symmetry in the structure of the animal.

#### Literature.

#### Agassiz, L.

'62. Contributions to the Natural History of the United States of America. Vol. IV. (Boston).

#### ALLMAN, G. J.

- '64. On the construction and limitation of genera among the Hydroidea. Ann. and Mag. of N. H., 3rd series, No. 77.
- '71. A monograph of the gymnoblastic or Tubularian Hydroids. (London.)
- '83-'88. Report on the Hydroids dredged by H. M. S. Challenger during the years 1873-76.

#### BRAUER, A.

'91. Ueber die Entstehung der Geschlechtsprodukte und Entwickelung von Tubularia mesembryanthemum. Zeit für wiss. Zool. Bd. 52.

#### CARLGREN, O.

699. Branchiocerianthus urceolus E. L. Mark, eine Hydroid? Zool. Aug. Bd. XXII. No. 581.

#### CHUN, C.

- '91. Coelenteraten, in Bronn's Klass. und Ord. des Tierreichs.
- '99. Die deutsche Tiefsee-Expedition 1898/1899. (Berlin).

#### CLARKE, S. F.

<sup>6</sup>76. Report on the Hydroids collected on the coast of Alaska and the Aleutian Islands, by W. H. Dall, U.S. Coast-Survey and Party, from 1871 to 1874.

#### Doflein, F.

- '96. Die Eibildung bei Tubularia; Z. für Wiss. Zool. Vol. 62.
- Grönberg, G.
  - '98. Beiträge zur Kenntniss der Gattung Tubularia. Zool. Jahrb. Bd. 11. Heft 1.

HAMANN, O.

'82. Der Organismus der Hydroid-Polypen. Jen. Zeit. Bd. 8. Vol. 8. (Jena).

HINCKS, TH.

'68. British Hydroid Zoophytes. 1868.

Von Koch, G.

- '73. Vorläufige Mittheilung über Cölenteraten. Jen. Zeit. Bd. 7. Mark, E. L.
  - '98. Preliminary report on Branchiocerianthus urceolus. Bull. of the Mus. of Comp. Zool. Vol. XXXII. No. 8.
  - '99. "Branchiocerianthus," a correction. Zool. Anz. Bd. XXII No. 590.

WEISMANN, A.

'83. Die Entstehung der Sexualzellen bei den Hydromedusen, 1883.

#### Explanation of Plate.

Pl. XIV.

The hydranth with the upper half and the lowest part of the hydrocaulus. Nat. size.

#### Pl. XV.

Reference letters: B, hypostomal region of the disc; Ect, ectoderm; End, endoderm; H upper, H' lower, cavity of the hydranth; ic, intercalated cord; mes, mesoderm: mt, marginal tentacle; p, peduncle of the gonosome: R, outer region of the disc, provided with the radial canals; rc, radial canals: t.b, human of the base of the marginal tentacle.

Fig. 1. Surface-view of a portion of the disc with gonosomes (p) and marginal tentacles (mt.). Nat. size.

a upper wall of the disc, b a part of the diaphragm in the hydranth.

Fig. 2. Longitudinal section through the outermost margin of the disc Zeiss  $a \times 4$ .

- Fig. 3. Cross-section of the upper wall of the lower cavity of the hydranth. Zeiss  $DD \times 2$ .
  - Fig. 4-9. Serial sections of the upper part of the disc. Zeiss  $a \times 4$ .

Fig. 4. Cross-section through the line 1-1 in Fig. 1.

Fig. 6. ,, ,, 3–3 ,,

Fig. 7. ,, ,, 4-4 ,

Fig. 8. ,. ,, 5–5

Fig. 9. ,, 6-6

Fig. 10. Cross-section of the radial canal and intercalated cord. Zeiss  $BB \times 2$ .

, ,

- Fig. 11. Cross-section of the marginal tentacle. Zeiss  $a_3 \times 2$ .
- Fig. 12. Longitudinal section of the same. Zeiss  $a_3 \times 2$ .
- Fig. 13. Terminal branches of a gonosome. Zeiss  $a \times 2$ .
- Fig. 14. Longitudinal section of a branchlet of the gonosome. Zeiss  $F \times 2$ . c.g. Central, l.g. lateral, globule.
  - Fig. 15. Central globule with nematocyst (n). Zeiss  $DD \times 4$ .
- Fig. 16. Lateral globule in which the bell-nucleus (b,n) and the endoderm-cup (enc.) are fairly well recognizable. Zeiss  $F \times 2$ .
- Fig. A part of the wall of the hydrocaulus with the wavy bands Nat, size.
- Fig. 18. Cross-section of the mesoderm in the hydrocaulus. Zeiss  $a \times 2$ . h.l. outer, longitudinally, c.l. inner, circularly, striated layer. x, spot corresponding to the wavy band.
- Fig. 19. Mesoderm of the hydrocaulus, macerated with caustic potash. Zeiss  $a \times 2$ .
- Fig. 20. Surface-view of the root and the sheath. Nat. size, ap, hair-like appendage; s, sheath.
- Fig. 21. Longitudinal section of the root figured in Fig 20. Zeiss  $a \times 2$ . o. ontgrowth of the endoderm; other letters as in Fig. 20.





# Mutual Relations between Torsion and Magnetization in Iron and Nickel Wires.

Bv

H. Nagaoka, Rigakuhakushi, Professor of Applied Mathematics,

and

**K. Honda**, Rigakushi, Post-graduate in Physics.

With Plate XVI.

The various effects of stress on the magnetization of ferromagnetic metals are of such a complex character that no simple relation seems to exist among them. The strains caused by magnetizing the ferromagnetics are of no less complex a nature, so that the co-ordination of these two classes of complicated phenomena is, up to the present, still a matter of doubt. Various isolated facts, such as the analogies between the change of magnetization by longitudinal pull and that of length by magnetization, the relation between the twist caused by the interaction of longitudinal and circular magnetizations and the circular or longitudinal magnetization produced by twisting a longitudinally or circularly magnetized wire respectively, were long considered as affording a clue to the explanation of these phenomena. So far

as we are aware, no attempt has yet been made to place all of these different phenomena on a common footing. Some time ago<sup>1</sup>, we hinted at the probable connections which exist between the twist caused by passing an electric current through a longitudinally magnetized wire and the change of volume and of length in ferromagnetic metals produced by magnetization. The said relation can also be extended to the explanation of other phenomena; namely, the transient current produced by twisting a magnetized wire and the longitudinal magnetization caused by twisting a circularly magnetized wire. It is our object in the present paper to show that these different phenomena can be linked together in a common bond.

### § 1. Twist produced by the interaction of circular and longitudinal magnetizations.

The subject was first studied by G. Wiedemann<sup>2)</sup> who established remarkable reciprocal relations with the longitudinal magnetization produced by twisting a circularly magnetized wire. Dr. Knott<sup>3)</sup> found that the direction of twist in iron is opposite to that in nickel; Bidwell<sup>4)</sup> afterwards discovered that the twist in iron is reversed in high fields and takes place in the same direction as in nickel. Unfortunately some of the experiments were undertaken with wires which were longer than that of the coil, so that the magnetization was far from being uniform. It will suffice for qualitative tests, but we can not hope for any

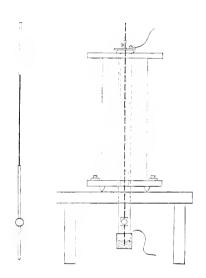
<sup>1)</sup> Nagaoka and Honda, this Journ. 13, p. 57, 1900; Phil. mag. 49, p. 341, 1900.

<sup>2)</sup> G. Wiedemann, Pogg. Ann. 103, p. 571, 1858; 106, p. 161, 1859; Elektricität, 3.

<sup>3)</sup> Knott, Trans. Roy. Soc. Edinb., **32** (1), p. 193, 1882/85; **35** (2), p. 377, 1889; **36** (2), p. 485, 1891.

definite quantitative results. The position of maximum twist in nickel shows a large difference in the present from the corresponding experiment by Dr. Knott.

The twist produced by longitudinal magnetization of a circularly magnetized wire was measured in the following way. To the extremities of an iron or nickel wire 21 cm. long were



brazed stout brass wires, and a light plane mirror was attached to the lower one. The end of the lower brass wire was dipped in a mercury pool, while the upper brass wire was clamped to a small tripod, which rested on the top of a magnetizing coil provided with hole, slot, and plane arrangement. One end of the accumulator was connected with the tripod, while the other was led to a mercury pool. The

wire hung vertically in the axial line of the coil, which was 30 cm. long and gave a field of 37.97 C.G.S. units at the centre by passing a current of one ampere. The vertical component of the terrestrial magnetic field was compensated by placing another coil in the interior of the magnetizing coil. The lower part of the wire to be tested was protected against air current by enclosing it in a wide brass tube with a small window, just where the reflecting mirror was attached. The twist was measured by scale and telescope method, by which the deflection of 0.3" per. cm. was easily read. The current was measured by Kelvin graded amperemeters, whose constants were

from time to time checked by means of an ampere balance. The experiment was conducted in the following manner:—

- 1. The circularly magnetizing current was kept constant, and the amount of twist measured by varying the longitudinally magnetizing current.
- 2. The longitudinally magnetizing current was kept constant, and the amount of twist measured by varying the circularly magnetizing current.

Before each experiment, care was taken to demagnetize the wire completely either longitudinally or circularly by passing an alternate current of gradually diminishing intensity.

Twist by varying the longitudinal field (Fig. 1).—The direction of twist in iron, so long as the longitudinal magnetizing field is not strong, is such that if the current is passed down the wire from the fixed to the free end and the wire is magnetized with north pole upwards, the free end, as seen from above, twists in the direction of the hands of a watch. By keeping the circular field constant, the amount of twist increases at first, till it reaches a maximum in a field of about 20 units; it then goes on diminishing till it ultimately changes the direction and continues to twist in the opposite direction with increasing field. The field at which the twist is reversed increases with the circularly magnetizing field. In nickel, the direction of twist is opposite to that in iron, but the general feature is similar to iron, the only difference being that even in strong longitudinal fields, the twist is not reversed. For wires of the equal thickness, the amount of twist in nickel is greater than that in iron—the maximum twist in iron wire of 1 mm. diam. by passing 6 amperes through it amounts to about 28" per cm., while with nickel wire of 0.83 mm. diam. under similar conditions, the maximum twist amounts to about 200."

Twist by varying the circular field (Fig. 2).—Here we notice a slight dissimilarity between iron and nickel. In iron, the twist increases with the strength of the circular field, if the longitudinal field remains constant. Such is also the case with nickel in moderate and strong fields. In low longitudinal fields, however, the twist does not continue to increase with the circular, but we notice a maximum as will be clear in the figure. There is great experimental difficulty in increasing the circular field, inasmuch as the heating of the wire becomes very great and thus materially deteriorates the result.

The hysteresis accompanying the cyclical change of the circular magnetization deserves special notice (see Fig. 3). If the longitudinal field be such that with the increase of the circularly magnetizing force, the twist reaches a maximum, the curve of twist goes below the former course on weakening the circular magnetization. The twist, however, goes on slowly increasing, till it crosses the on-curve and then reaches a maximum, whence it gradually diminishes and ultimately vanishes in negative field. The course after passing this point is exactly the reverse of that already described. The character of twist is exactly the same for iron as for nickel, when we take the opposite character of twist into account. The nature of the hysteresis is nearly the same when the longitudinal magnetizing field is made to vary, while the circular field remains constant.

The results thus far obtained are in accordance with the experiments of Wiedemann and Knott: we have only to notice the discrepancy as regards the position of maximum twist in nickel. In Dr. Knott's experiment, the said point occurs in tolerably high field, while in the present experiment, it occurs nearly in the same field as in iron. It may partly be due to the

difference in the method of measuring the twist and partly to the non-uniformity of the field, as was often the case in most of the older experiments.

The observed angles of twist in iron and nickel are exhibited in the following tables, where C denotes the longitudinal current per sq. mm. in amperes, H the field strength in C.G.S. units and  $\tau$  the angle of twist per cm. expressed in seconds.

Circular Field being Constant.

Iron wire: diam.=0.98 mm.

C=	= 1.06	C = 3.38		C = 6.95	
Н	τ	Н	τ	Н	τ
5.6	18.6"	5.6	21.9''	5.6	16.4"
12.4	21.7	11.3	35.6	11.3	30.0
26.0	13.1	22.6	32.8	22.6	36.4
41.3	7.9	36.2	24.7	49.2	27.8
90.5	2.5	79.2	13.7	78.0	19.7
112.0	1.6	97.3	10.7	96.2	16.4
215.0	-0.5	191.0	6.0	132.0	8.2
492.0	- 1.9	442.0	2.7	455.0	2.5

Nickel wire: diam.=0.83 mm.

C=	C = 2.45		C = 4.33		=6.05
Н	τ	Н	τ	Н	τ
4.9	- 94.1"	4.9	- 97.6"	4.9	- 88.6"
11.2	-137.0	11.2	-164.4	11.2	-159.8
24.3	-125.5	24.3	-179.4	24.3	-196.4
38.8	-102.1	38.8	-156.6	38.5	-182.6
53.6	- 85.1	53.4	-134.1	53.2	-159.2
84.0	<b>-</b> 62.9	83.8	-101.8	84.0	-119.7
102.2	-54.5	102.2	- 88.0	103.4	-101.8
225.0	-31.7	222.0	-47.9	226.0	- 59.3
415.0	- 19.6	389.0	- 29.8	414.0	-40.2

Longitudinal Field being Constant.

Iron Wire: diam. = 1.05 mm.

H=	2.67	H = 5.92 $H = 19$		9.38	
C		C	=	C	τ
0.35	0.3"	0.58	0.3"	0.40	0.0"
0.80	1.1	1.03	0.6	0.89	0.0
1.06	2.0	1.73	2.0	1.36	0.0
1.80	3.7	2.20	3.0	1.92	0.3
2.22	5.7	2.96	5.1	2.66	0.4
3.04	8.8	3.55	6.2	3.48	0.6
4.35	14.2	4.78	9.1	4.88	1.4
6.79	21.5	6.73	13.6	6.25	2.5
7.52	23,2	7,34	14.7	7.16	2.8

Nickel wire: diam. = 0.83 mm.

Н=	= 5.7	H = 39.7		H=	-96 <b>.</b> 3
$^{-}\mathrm{C}$	τ	C	7	$^{\mathrm{C}}$	τ
1.03	22.1"	0.95	22.6"	0.81	8.5"
1.89	48.1	2.06	59.3	1.76	19.9
2.82	70.2	3.34	101.4	2.82	32.3
4.50	102.0	4.10	123.6	4.29	50.5
7.25	121.2	5.90	153.6	6.22	73.2
9.40	116.4	7.95	172.8	8.65	99.6
11.60	107.4	11.40	178.8	11.50	127.5
13.38	99.0	13.10	176.4	13.22	141.6

## § 2. Circular magnetization produced by twisting a longitudinally magnetized wire.

By twisting a longitudinally magnetized ferromagnetic wire, circular magnetization is developed. If, therefore, two ends of the wire are connected by a conducting wire, a transient current due to the circular magnetization appears in the circuit at the moment when the twist is applied. Some years ago,

one<sup>1)</sup> of us investigated the transient current for iron and nickel wires. It was then found that the current due to twisting was opposite in direction in these two metals and that it reached a maximum in moderate fields. As the magnetizing current was not very strong, no conclusive measurements were made as regards the nature of the transient current in strong fields. In order to make this point clear and see if any intimate relation with the Wiedemann effect could be traced, fresh experiments were undertaken by the same method as before. We have to notice that the ferromagnetic wire was so placed in the axial line of the magnetizing coil that it lay in nearly uniform field.

Some of the measurements of the transient current for iron and nickel wires are given in the following table and in Fig. 4.

Iron	Iron wire: diam. = 1.33 mm.		Nickel wire: diam. = 1.09 mm.					
	ler	gth = 20.9	0 cm.		len	gth = 20.8	gth = 20.80  cm.	
U =	=1.5°	$\theta =$	:50°	$\theta =$	:15°	$\theta =$	50°	
Н	$0.1 \times 10^{\circ}$	$\sim 11$	$Q \times 10^{s}$	11	$Q \times 10^{\circ}$	$\mathbf{H}$	$Q \times 10^{s}$	
3.2	25.4	3.2	30.7	1.3	- 5.8	1.1	- 2.5	
5.3	29.0	4.8	33.6	3.7	-7.1	2.7	<b>-</b> 6.2	
15.6	24.9	15.8	42.0	16.0	-17.7	5.3	-18.5	
32.6	16.3	30.5	38.4	33.7	-21.2	10.2	-27.1	
.545	10.9	48.6	30.5	53.4	-21.5	25.6	-38.0	
87.4	6.4	86.0	18.5	81.2	-20.1	44.9	-41.7	
120.8	2.5	139.4	8.1	115.4	-19.2	67.6	-40.7	
165.6	1.5	183.8	3.7	157.0	-16.4	107.4	-40.0	
242.0	-0.1	327.6	-3.2	213.7	-12.6	160.2	-35.7	
495.4	-1.7	447.4	<b>- 5.</b> 0	319.6	-10.3	241.5	-29.1	
650.2	-2.9	711.0	-5.1	530.3	-6.5	316.4	-26.7	
908.0	- 3.3	959.0	-5.2	703.0	- 5.4	699.8	-13.9	
1298.0	- 3.1	1521.0	-4.7	1214.0	<b>-</b> 3.9	1150.0	-10.6	
1790.0	- 2.5	1872.0	- 4.4	1815.0	<b>-</b> 2.0	1853.0	<u>- 8.9</u>	

<sup>1)</sup> Nagaoka, Journ. Sei. Coll., Tōkyō, 4, p. 323, 1891.

Here  $\theta$  denotes the angle of torsion and Q the time-integral of the transient current expressed in C.G.S. units. The resistance of the whole circuit was 4.5 ohms. The nickel wire here used was made of the same specimen as the nickel prism used in our former experiments.

As is well known, the direction of the transient current, and therefore that of the circular magnetization, is opposite in iron and nickel. The current for constant amount of twist increases with the strength of the longitudinal field; it, however, soon reaches a maximum, whence it gradually diminishes. In nickel, the transient current attains asymptotic values in strong fields without changing its direction, while in iron, it is reversed in a field of about 200 C.G.S. units, when the twist is small. The increase after the reversal is not pronounced, but becomes finally asymptotic.

### § 3. Longitudinal magnetization produced by twisting a circularly magnetized wire.

The longitudinal magnetization produced by twisting a circularly magnetized wire presents the same character as the transient current above described. The experiment is very difficult on account of the heating of the wire. To avoid the rise of temperature, the iron or nickel wire to be tested was covered with urushi (Japan lac) which has the special property of being a very good insulator while, at the same time, the melting temperature is comparatively high. The wire thus insulated was stretched in the axial line of a secondary coil whose diameter was 1.5 cm. and whose total number of turns was 540, and the current of cold water was kept flowing about it to keep the temperature

of the wire uniform. Thus maintaining the electric current in the wire constant, it was twisted and the induced current in the secondary circuit due to the longitudinal magnetization thereby developed was measured by the ballistic method.

Some of the results of observations are given in the following table and graphically shown in Fig. 5.

Iron	wire: dian lengt	h = 0.88 $h = 20.74$		Nickel	n = 0.965  mm. $th = 20.94  cm.$	
$\theta =$	:15°	$\theta =$	50°	$\theta =$	15°	$\theta = 50^{\circ}$
C	$Q \times 10^9$	C	$Q \times 10^9$	C	$Q \times 10^9$	$C = Q \times 10^9$
0.21	0.7	0.21	16.1	0.21	- 45	0.14 - 133
0.85	3.7	0.69	45.3	0.66	-111	0.23 - 294
1.53	20.5	1.49	89.1	1.56	-205	0.90 - 397
2.36	31.4	2.19	111.0	2.40	-239	2.05 - 448
3.93	36.5	3.27	125.6	3,35	-256	2.87 - 450
4.72	33.6	4.65	124.2	4.36	-265	4.42 - 452
6.55	29.2	5.91	115.4	5.86	-272	7.37 - 448
7.89	21.9	8.29	109.6	7.95	-269	10.34 - 431
12.82	13.9	12.48	86.2	10,89	-256	15.33 - 407
19.01	10.9	17.08	65.7	14.07	-243	20.85 - 397
24.29	5.8	24.37	51.8	20.18	-219	23.04 - 378
28.64	5.8	29.14	43.2	26.46	206	26.13 - 362

C denotes the total current through the wire expressed in amperes;  $\theta$  and Q have the same meanings as before.

As will be seen from the figure, the quantity of induced electricity in the secondary circuit, and therefore the longitudinal magnetization developed, by twisting a circularly magnetized iron wire attains a maximum, when the mean circular field is about 10 units. It then decreases, but in spite of the

constant stream of water, the heating due to electric current prevented the experiment from being pushed to the point where the direction of the current is reversed. However, to judge from the course of the curve, the tendency is such that there is a reversal. In nickel, the direction of the induced current is opposite to that in iron, and the total quantity of the current attains a maximum, whence it continually diminishes, but not to such an extent that the current ultimately changes its direction.

These experiments show that the twist produced by the combined action of the longitudinal and circular magnetizations, the circular magnetization produced by twisting a longitudinally magnetized wire, and the longitudinal magnetization caused by twisting a circularly magnetized wire are characterized by having various peculiarities, which are common to all of them. This can not be a mere chance coincidence; we shall have to ascribe these allied phenomena to the same common cause.

In the experiments of this and the last paragraphs, we were assisted by Mr. S. Shimizu, a post-graduate in physics, to whom our best thanks are due.

## § 4. Theory.

As already remarked in our last paper on magnetostriction, Kirchhoff's theory can be extended to the study of the relation between torsion and magnetization, exactly, in the same manner as was done by Maxwell and Chrystal to explain the Wiedemann effect. There we found that the mean circular magnetization called into play by twisting a ferromagnetic wire of radius R through angle  $\omega$  amounts to

$$-\frac{1}{3}\omega k'' IIR. \qquad (A)$$

in field H, and that the mean longitudinal magnetization caused by twisting a ferromagnetic wire carrying an electric current C amounts to

$$-\frac{1}{2}\omega k'' C. \qquad (B)$$

The reciprocal relation between these two phenomena is thus apparent at a glance. We shall next show how the same phenomena are reciprocally connected with the torsion produced by the interaction of the longitudinal and circular magnetizations.

The stress components in a magnetic medium as given by Kirchhoff are as follows:

$$\begin{split} X_{x} &= -\left(\frac{1}{4\pi} + k + \frac{k''}{2}\right)a^{2} + \frac{1}{2}\left(\frac{1}{4\pi} + k - k'\right) \left(a^{2} + \beta^{2} + \gamma^{2}\right). \\ Y_{y} &= -\left(\frac{1}{4\pi} + k + \frac{k''}{2}\right)\beta^{2} + \frac{1}{2}\left(\frac{1}{4\pi} + k - k'\right) \left(a^{2} + \beta^{2} + \gamma^{2}\right). \\ Z_{z} &= -\left(\frac{1}{4\pi} + k + \frac{k''}{2}\right)\gamma^{2} + \frac{1}{2}\left(\frac{1}{4\pi} + k - k'\right) \left(a^{2} + \beta^{2} + \gamma^{2}\right). \\ Y_{z} &= Z_{y} &= -\left(\frac{1}{4\pi} + k + \frac{k''}{2}\right)\beta\gamma. \\ Z_{x} &= X_{z} &= -\left(\frac{1}{4\pi} + k + \frac{k''}{2}\right)\gamma\alpha. \\ X_{y} &= Y_{x} &= -\left(\frac{1}{4\pi} + k + \frac{k''}{2}\right)\alpha\beta. \end{split}$$

Taking the axis of z in the axial line of the wire, and two other axes in the plane perpendicular to it, we see that the component

magnetic forces in a longitudinally magnetized wire traversed by an electric current are

$$a = -h \sin \theta, \qquad \beta = h \cos \theta, \qquad \gamma = H,$$

where h denotes circular field given by

$$h = \frac{2Cr}{R^2},$$

C being the current, r the distance of the point from the axis of the wire, R the radius, and  $\theta$  the angle between r and the axis of x.

The stress components in ferromagnetic medium acted upon by the forces above specified are given by

$$\begin{split} X_x &= - \left( \frac{1}{4\pi} + k + \frac{k''}{2} \right) h^2 \sin^2 \theta + \frac{1}{2} \left( \frac{1}{4\pi} + k - k' \right) \left( H^2 + h^2 \right), \\ Y_y &= - \left( \frac{1}{4\pi} + k + \frac{k''}{2} \right) h^2 \cos^2 \theta + \frac{1}{2} \left( \frac{1}{4\pi} + k - k' \right) \left( H^2 + h^2 \right), \\ Z_z &= - \left( \frac{1}{4\pi} + k + \frac{k''}{2} \right) H^2 + \frac{1}{2} \left( \frac{1}{4\pi} + k - k' \right) \left( H^2 + h^2 \right), \\ Y_z &= Z_y = - \left( \frac{1}{4\pi} + k + \frac{k''}{2} \right) h \ H \cos \theta, \\ Z_x &= X_z = \left( \frac{1}{4\pi} + k + \frac{k''}{2} \right) h \ H \sin \theta, \\ X_y &= Y_x = \left( \frac{1}{4\pi} + k + \frac{k''}{2} \right) h^2 \sin \theta \cos \theta. \end{split}$$

The moment about the axis of the wire is given by

$$\begin{split} N &= \int \int \left( Z_y \, x - Z_x \, y \right) \, dx dy, \\ &= - \int \int \left( \frac{1}{4\pi} + k + \frac{k''}{2} \right) h \, H \, r \, dx dy, \\ &= - \frac{2}{R^2} \left( \frac{1}{4\pi} + k + \frac{k''}{2} \right) C H \, \int_{\bullet}^{R} \int_{\sigma}^{2\pi} dr d\theta, \\ &= - \pi \left( \frac{1}{4\pi} + k + \frac{k''}{2} \right) C H R^2. \end{split}$$

Since  $-\frac{1}{4\pi}$  and k are very small compared with k'', the torsional couple twisting the wire amounts nearly to

$$-\frac{\pi k''}{2}CHR^2 = -\frac{k''CH}{2} \times Cross \ section. \tag{C}$$

Since the amount of torsion of a cylindrical wire by a given couple is inversely proportional to the fourth power of its radius, it is evident that for given longitudinal current and field, the angle of twist is inversely proportional to the square of the radius. This inference was approximately verified in the present experiments.

In deducing the three formulae (A), (B), (C), we can not, strictly speaking, put k'' outside the sign of integration, because the strain coefficient depends on the field strength, which is not uniform in a wire traversed by electric current. Hence in these formulae, we shall have to use a mean value to obtain a close approximation.

The mutual relations between twist and magnetization are embodied in the three formulæ above given. There we notice that the strain coefficient k'' determines the nature of the three different phenomena studied in the above experiments. The fact that the coefficient k'' is principally determined by the elongation in the ferromagnetic metal accounts for the close analogy between the said phenomena and the elongations due to magnetization. As the above result imports, the analogy is not exact, inasmuch as the elongation is also affected by terms depending on k', which depends mostly on the change of volume.

In order to test the consequences of the theory as regards the twist produced by the joint action of circular and longitudinal magnetizations, we have calculated the twist by assuming the

values of k'', calculated from the changes of volume and of length in iron and nickel ovoids. Graphically represented (Fig. 6.), the fields of maximum twist by calculation coincide nearly with that given by experiments, and the reversal of twist in iron takes place in low fields as actually found by observation. The quantitative differences are, however, tolerably large in iron, but in nickel the amount of twist is nearly coincident with the experimental values. Calculating, in the same manner, the quantity of the transient current produced by twisting longitudinally magnetized wires, we find a close coincidence between the experimental and theoretical values in nickel, but the difference is tolerably large in iron. In using the strain coefficients, we must always bear in mind that these values are widely different according to the nature of the specimen; especially with wires, we are not sure of its being magnetically isotropic. The apparent discrepancy would probably be lessened, if we could measure the twist as well as the strain coefficients on the same specimen. The remarkable qualitative coincidence as regards the existence of maximum twist and its reversal in iron are convincing proofs that the theory, so far as we know at present, admits of connecting various experimental facts in a common bond.

As regards the mutual relations among the three different phenomena above enumerated, it will suffice to state that several of them have already been noticed by G. Wiedemann in his researches on the relation between torsion and magnetism. He especially studied the relation between permanent torsion and the effect of magnetizing the twisted wire. The principal object of his researches was to expose the different aspects of the phenomena involved in the relation between torsion and magnetization in order to bring to light his ingenious theory of rotatory molecules.

Elegant as it at first sight appears to be, Wiedemann's theory abounds with hypotheses which we are not always warranted in making.

In his work on the applications of dynamics to physics and chemistry, J. J. Thomson has propounded a new method of investigating the mutual relations between the effects of various physical agencies. He showed that the existence of a certain phenomenon involves as a natural consequence that of another reciprocating with it. As an application of his method, he showed that if the wire be twisted by the interaction of longitudinal and circular magnetizations, a transient current will be produced simply by twisting a longitudinally magnetized wire and a longitudinal magnetization will be developed by twisting a circularly magnetized wire.

The peculiar feature of Kirchhoff's theory lies in the simple and natural way of elucidating the relations between the various kinds of strain caused by magnetization and the effects of stress on magnetization. Just as we can study the various elastic behaviour of isotropic bodies by knowing the bulk- and stretch-moduli, we have to deal, in Kirchhoff's theory, with the strain coefficients k' and k'' which play the  $r\delta les$  of different moduli in the theory of elasticity.

The reciprocal relations between the strain caused by magnetization, and the effects of stress on magnetization, as found by actual experiments, will be found to be of paramount importance in arriving at a correct theory of magnetostriction. The strain accompanying the magnetization of ferromagnetic metal will be determined, when we know the effects of stress on magnetization and vice versa. As regards the relations between twist

and magnetization, we may conveniently place them under the following parallel statements:

#### Strains produced by magnetization.

- (a)—(Experiment and theory). A longitudinally magnetized wire is twisted by circular magnetization.
- (b)—(Experiment and theory). A circularly magnetized wire is twisted by longitudinal magnetization.
- (c)—(Experiment and theory). Up to moderate fields, the twist produced by the longitudinal and circular magnetizations of an iron wire is opposite to that in nickel.
- (d)—(Experiment and theory). The twist due to longitudinal magnetization of a circularly magnetized iron or nickel wire reaches a maximum in low fields.
- (e)—(Experiment and theory). In strong fields, the twist due to longitudinal magnetization of a circularly magnetized iron wire is reversed and takes place in the same direction as in nickel.

#### Effects of Stress on magnetization.

- (a')—(Experiment and theory) Twisting a longitudinally magnetized wire gives rise to circular magnetization.
- (b')—(Experiment and theory). Twisting a circularly magnetized wire gives rise to longitudinal magnetization.
- (c')—(Experiment and theory). Up to moderate fields, the transient current, or the longitudinal magnetization produced by twisting a longitudinally or circularly magnetized wire respectively, is opposite to that in nickel.
- (d')—(Experiment and theory). The transient current produced by twisting a longitudinally magnetized iron or nickel wire reaches a maximum in low fields.
- (e')—(Experiment and theory). In strong fields, the direction of the transient current produced by twisting a longitudinally magnetized iron wire is reversed and is in the same direction as in nickel.

In his paper on the principle of least action, Helmholtz<sup>1)</sup> has placed the reciprocal relations of a dynamical system under three heads. Denoting the generalized co-ordinates, the veloci-

<sup>1)</sup> Helmholtz, Crelle's Journal 100, p. 137, 1886; Abh., 3, p. 203, 1895.

ties, the accelerations and the forces by p's, q's, q's, and P's, the relations are generally expressible by the equations

$$(1) \quad \frac{\partial P_a}{\partial \rho_b} = \frac{\partial P_b}{\partial \rho_a},$$

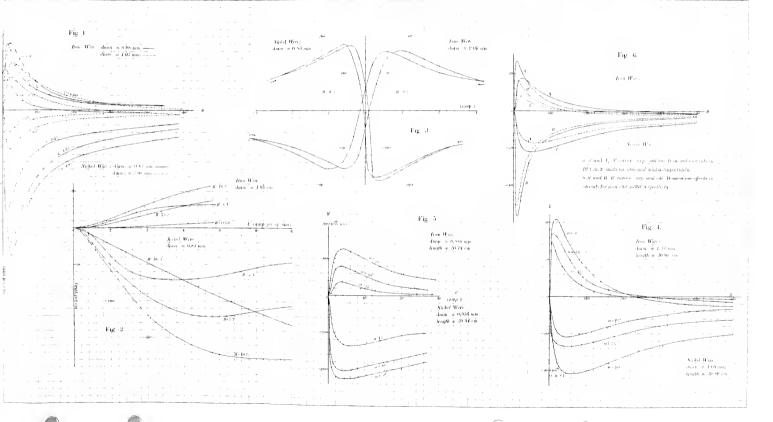
$$(2) \quad \frac{\partial P_a}{\partial q_b} = - \quad \frac{\partial P_b}{\partial q_a} \ ,$$

$$(3) \quad \frac{\partial P_a}{\partial q'_b} = \frac{\partial P_b}{\partial q'_a}.$$

It will be easily seen that the relations above cited belong to case (2).

The greatest difficulty that we encounter in establishing the relations between the effects of stress on magnetization and the strain caused by magnetization lies in the great difference of strain coefficients according to the nature of the specimen. If all the experiments be performed in a proper manner on one and the same specimen of ferromagnetic metals, we may feel assured of being able to discern the true merits of the theory, or to detect its various defects, not only from qualitative points of view, but also in various quantitative details.







### The Interaction between Sulphites and Nitrites.

By

Edward Divers, M. D., D. Sc., F. R. S., Emeritus Prof.,

and

Tamemasa Haga, D. Sc., F. C. S., Professor, Tōkyō Imperial University.

The present paper gives an account of a series of experiments, the results of which seem to leave no room for doubt as to the truth of the following propositions respecting the sulphonation of nitrites:—(1) normal sulphites are inactive upon nitrites; (2) pyrosulphites are not active to their whole extent upon nitrites; (3) pyrosulphites are active in their entirety upon nitrous acid or its equivalent of nitric oxide and nitric peroxide (nitrous fumes); and (4) sulphurous acid and nitrous acid or the oxides and water equivalent to them interact of themselves and in such a way that the base of the sulphite, that may be used in place of the sulphurous acid, is needed only to preserve from hydrolysis the products of their interaction. Concerning these assertions we would point out that the first directly contradicts the conclusions drawn by other workers from their experiments; the

second is novel like the first, the facts on which it is based having been misunderstood; the third has indeed been already enunciated, but not with intention to limit it to a strict interpretation, and only theoretically, without any experimental treatment of it; and, lastly, the fourth has been also made before and upon the basis of experiment, but experiment quite inadequate to justify it.

The establishment of these propositions, taken along with what we have already published as to the constitution of Fremy's salts will then allow of the further assertion being made, that the interaction of nitrous acid with a pyrosulphite results in their conversion into a two-thirds normal hydroximidosulphate (and water), and that all the other sulphazotised salts are secondary products simply derived. It is thus established that the only interaction between sulphites and nitrites is one of the greatest simplicity, instead of being full of complications, as hitherto believed.

## I.—a. A Normal Sulphite inactive upon a Nitrite.

Dipotassium or disodium sulphite is quite inactive upon a nitrite. In establishing this fact we have mixed solutions of normal sulphite and nitrite in proportions varying in different experiments, and left them in closely corked flasks, almost full, for days and for weeks. No change has ever happened. When coloured with rosolic acid, a drop of dilute acid would at any time, as at first, discharge the colour. Had any action occurred, alkali hydroxide must have been generated (as to the possibility of which see sect. II. b.). A portion of the solution to which had been added a drop of dilute sulphuric acid reacquired the pink colour of the rosolic acid when left to stand for some time

—the minute quantity of pyrosulphite which the acid had produced having slowly interacted with the nitrite, but when that was used up, no more action occurred and, at any time added one drop of dilute acid would again remove the colour of the solution.

No further proof of the activity upon a nitrite of a normal alkali sulphite is wanted, but additional evidence of the fact is readily obtainable. Thus, in the case of the potassium salts, while the action of pyrosulphite upon nitrite shows itself (except in cases of high dilution) by the formation of the insoluble nitrilosulphate, no separation of this salt occurred in the above experiments. Again, barium chloride, added, at any time, to the mixed and to litmus very alkaline solution of normal sulphite and nitrite, precipitated all the sulphur as sulphite (with also a very little sulphate) and left the nitrite in solution, neutral to litmus. Had sulphazotised salts been present, precipitation of the sulphur would have been incomplete, and the mother-liquor would have preserved a strong alkalinity to litmus and when acidified soon have deposited barium sulphate in the cold and at once if boiled.

Fremy, Claus, and Raschig all believed in the activity of dipotassium sulphite upon potassium nitrite, although the last named chemist recognised the value also of the pyrosulphite, as Berglund had done before him. Fremy apparently used sulphite neutral to litmus and took it to be the normal salt, and Claus certainly did so. Since, therefore, they used sulphite which was, for the most part, pyrosulphite, no evidence upon the point in question can be gathered from their work. We would account for Fremy's finding sodium sulphite inactive upon sodium nitrite, otherwise inexplicable, by assuming that the solution of sodium sulphite which he tried happened to contain no pyrosulphite.

Claus's statement that some potassium sulphite neutral to litmus was as active upon nitrite, after he had added potash in 'excess' to it, as it was before only requires us to assume that the excess spoken of was large enough to give the solution a markedly alkaline action upon litmus and yet small enough to leave much pyrosulphite unchanged.

## I.—b. Potassium Hydroxide not a Factor in the Formation of the Sulphazotised Salts.

That a normal sulphite, potassium or sodium, remains still inactive upon nitrite, when alkali hydroxide is added, was ascertained by leaving the three substances together in solution in a closed flask for some time, as in the experiments where no hydroxide was present, and then precipitating with barium chloride after addition of ammonium chloride, and finding no sulphur compound left in the filtrate. (Ammonium chloride prevents precification of hydroximidosulphate, this Journal 7, 48, 69).

On the many occasions we have had to prepare sulphazotised potassium salts by submitting solutions of nitrite and hydroxide to the action of sulphur dioxide, taking care to keep the solution briskly agitated, we found that, even in ice-cold solutions, precipitation of these very sparingly soluble salts only began from the point at which there was no more hydroxide left, and then went on freely until the solution had become neutral to laemoid paper. In proportion as the hydroxide disappeared, sulphite became abundant, whilst from the time that the replacement of hydroxide by sulphite was complete, the quantity of sulphite steadily decreased as the sulphazotised salts formed, sulphazotised sodium salts being very soluble no preci-

pitation occurs during their preparation, and with these, therefore we made an experiment to determine quantitatively what happens up to the point when the last portion of hydroxide disappears, a point indicated by rosolic acid losing its pink colour.

Washed sulphur dioxide was sent in a steady stream into a solution of 11.21 grams real sodium hydroxide and 19.34 grams sodium nitrite in about 198 grams water, kept in active motion and immersed in ice. The sodium compounds were in molecular proportions; more nitrite would not have mattered. In a short time a 10 cc. pipetteful was removed; soon after a second; and not very long after a third one, just when the pink colour of the rosolic acid present had disappeared. The three portions were treated alike. Each was mixed with excess of solutions of ammonium and barium chlorides and the precipitate filtered off, oxidised to sulphate, washed with dilute hydrochloric acid, and weighed as barium sulphate. The ammonium-chloride filtrate was evaporated to dryness, during which operation all the sulphur of the sulphazotised salts was converted to sulphate by the nitrite and ammonium chloride. The soluble salts were washed out with dilute hydrochloric acid, and the barium sulphate collected and weighed. The solution of salts removed from the barium sulphate was concentrated and then heated under pressure for hours, after which it was found to be still clear and therefore free from sulphate, which must have formed by hydrolysis had any sulphazotised salt escaped decomposition during the evaporation with nitrite and ammonium salts.

We give the quantities of sulphur dioxide found in each pipetteful as sulphite and as sulphazotised salts, and also state these quantities as parts per hundred of the total sulphur dioxide which had entered it.

Sulphur dioxide 1st 10 cc. 2nd 10 cc. 3rd 10 cc. Sulphite .0662 grms. = 96.6% .1204 grms. = 96.5% .3996 grms. = 91.5% Sulphazot. .0023 , = 3.4% .0047 , = 3.5% .0365 , = 8.5%

It will be seen that all but 3.5 per cent. of the sulphur dioxide entering the solution in the early stages of the experiment remained in the form of sulphite, and that even up to the time when the last of the hydroxide had been consumed, all but 8.5 per cent. of the total sulphur dioxide was in the state of sulphite. That it must be impossible to prevent all temporary local excess of sulphur dioxide will be at once admitted, as also that it must be difficult in the later stages to keep down this local excess to very narrow limits. Therefore it will seem in the highest degree probable, if not certain, from this experiment that sulphur dioxide, equally with normal sulphite, does not act upon nitrite in presence of alkali hydroxide.

Now Fremy believed that potassium hydroxide helps the formation of sulphazotised salts and endeavoured, accordingly to keep some of it always present when passing sulphur dioxide into a solution of potassium nitrite. This he did by adding it occasionally during the process, and to such an extent that at the end of the operation the mother-liquor of his salts was always not merely alkaline but eaustic and destructive to filter paper. Claus did not go so far as to believe that the potash exercised any specific influence upon the action between the sulphur dioxide and the nitrite, but, agreeing with Fremy as to its value in precipitating and preserving the sulphazotised salts, he adopted the precaution to stop passing in sulphur dioxide when the alumina contained in the potassium hydroxide began to precipitate, since this occurs while the solution is still strongly alkaline to litmus. Raschig, in attempting to prepare Fremy's

sulphazate, also used precipitation of alumina as the indication to stop the process while yet alkali remained.

Thus, then, it would seem, Fremy, Claus, and Raschig, the last in less degree, have all prepared sulphazotised salts without difficulty, under conditions which we pronounce to be incompatible with their production. To remove this apparent contradiction in results it is sufficient to assume, for one thing, that, in Fremy's way of working, success followed only because, temporarily and locally, the point of saturation of the alkali was reached and exceeded again and again where the gas entered the solution,—a state of things, never avoidable altogether, above all at the time when the potassium hydroxide is nearly exhausted. There is nothing to show that, to check this, he kept his solution well agitated. Secondly, we can assume, with great probability that his solution often lost its alkalinity between the additions of the hydroxide which he made. Working as we believe he actually did, we have found it easy to get results such as his. So far as Claus and Raschig followed Fremv's method, their results are equally open to objection, while it is to be remarked of their alumina indicator that, not only is normal sulphite alkaline to litmus but, as we have found, any aluminium present is precipitated as hydroxide just when the sulphur dioxide has converted all alkali into normal sulphite. They will, therefore in their experiments have preserved none of the alkali unchanged and most probably have generated also some pyrosulphite. There is besides indirect evidence in Claus's work that normal sulphite is either inactive or only very slowly active upon nitrite, for when, having taken no excess of this salt, he stopped the process just after precipitation of alumina, much of this nitrite remained in the solution, while, as we have just pointed out, much normal

sulphite must also have been present. The two salts were, therefore, together in solution unchanged. Raschig, too, found that sulphite and nitrite are inactive upon each other when in presence of potassium hydroxide dissolved in only its own weight of water.

# II.—a. Even Pyrosulphite only active upon a Nitrite till it has become Normal Sulphite.

Pyrosulphite, neutral to lacmoid paper and containing therefore, neither sulphurous acid nor normal sulphite, freely sulphonates nitrite, but is far from being all consumed in the process, as it has been represented to be by Claus, Berglund, and Raschig. Quantitative experiments have shown us that, when pyrosulphite is left in solution with excess of nitrite in a closed vessel for a considerable time, about one-third of the sulphite remains inactive by becoming converted into the normal salt, separable, as in other eases, from the sulphazotised salts by precipitation with barium chloride in presence of ammonium chloride. From this it follows that 3 mols. pyrosulphite are needed to convert 2 mols. nitrite into hydroximidosulphate (this Journal, 7, 19) and not 2 mols. only, as had been supposed. The third mol. sulphite remains unavoidably in the solution but all the nitrite sulphonated:  $-2\text{NaNO}_2 + 3\text{Na}_2\text{S}_2\text{O}_5 + \text{OH}_2 = 2\text{Na}_3\text{HNS}_2\text{O}_7 + 2\text{Na}_2\text{SO}_3$ using less pyrosulphite, some nitrite remains at the end along with normal sulphite. That sodium pyrosulphite is not easily all used up in sulphonating sodium nitrite was observed by Raschig.

Not only hydroximidosulphate but a little nitrilosulphate is formed when a pyrosulphite acts upon a nitrite, but this need never be enough, with ordinary care, to cause much less than one-third of the sulphite to remain inactive. If excess of pyrosulphite is used the interaction appears to be—NaNO<sub>2</sub>+2Na<sub>2</sub>S<sub>2</sub>O<sub>5</sub>= Na<sub>3</sub>NS<sub>3</sub>O<sub>9</sub>+Na<sub>2</sub>SO<sub>2</sub>, but we have not made any quantitative determination of the sulphite remaining, the qualitative evidence being sufficient.

The interaction between pyrosulphite and nitrite proceeds at first very rapidly and with great elevation of temperature, but, when the temperature is kept down by cooling, soon slows down, so as to require many hours for its completion. The normal sulphite seems here to inhibit the action of the pyrosulphite, just as does the salt of a weak acid inhibit the action of that acid, an effect now well recognised. This consideration points to the propriety of looking upon the passage of pyrosulphite to normal sulphite as its action as an acid upon the nitrite, and not as the yielding up of half of its sulphurous acid for the sulphonation of the nitrite, the interactions being  $2NaNO_2 + Na_2S_2O_5 + OH_2 = 2HNO_2 + 2Na_2SO_3$ , and then  $2HNO_2 + 2Na_2SO_5 = 2Na_2HNS_2O_5$  (see section III a).

## II.—b. Alkali not produced in the Sulphonation of a Nitrite.

One of the most remarkable things, according to Claus, is the production of potassium hydroxide by the formation of Fremy's salts through the agency of a sulphite. He explained this production by the equation— $KNO_2+2K_2SO_3+2OH_2=K_2HNS_2O_7+3KHO$ . Such an equation was also published by Berglund (Lunds Univ. Årskr. 1875, 13, 14). Raschig gave the same equation for results obtained by himself and, in order to express

other results, gave also the equation— $NaNO_2+2NaHSO_3=Na_2HNS_2O_7+NaHO$ . Finding also, and again in agreement with Claus, that dipotassium hydroximidosulphate does not combine at once or even at all with potassium hydroxide, he argued that this salt cannot have a similar constitution to that of Fremy's 'basic' sulphazotate because potassium hydroxide is produced along with it instead of being combined with it as Fremy's 'basic' sulphazotate.

Now, all this is wrong in fact on the part both of Claus and Raschig, as we have already shown (this Journal 7, 30) or here show in other sections of the present paper, except as to the generation of alkali hydroxide, which we now proceed to deal with. Claus's emphatic statement, supported as it is by Berglund and by Raschig, that potassium hydroxide is formed when a sulphite meets a nitrite in solution, rests upon no other evidence than what we now set down in full, recalling the fact (section I, a.) that between the normal sulphite and nitrite there is really no activity of any kind. A solution of sulphite made neutral to litmus and a solution of nitrite of either potassium or sodium become hot and strongly alkaline to litmus when mixed together, and then contain much hydroximidosulphate and nitrilosulphate, both neutral to litmus, which soon crystallise out if they are the potassium salts. That is all these chemists had as evidence for the production of the hydroxide; let us add to these facts that addition of excess of barium chloride removes all the alkalinity. It follows, since pyrosulphites are a little acid to litmus and normal sulphites are very alkaline to it, that the phenomena depended upon offer no grounds whatever for the belief that alkali hydroxide is produced. Except by the use of lime, baryta, or other base, there is, we believe, only one way by

which potassium hydroxide can be generated from potassium sulphite and that is one made known by us, namely, treatment of the sulphite first with nitric oxide and then with alcohol and water (this Journal, 9, 106).

#### III.—a. A Pyrosulphite all active upon Nitrous Acid.

As remarked at the end of section II. a., a pyrosulphite appears to act as an acid upon the nitrite and then sulphonates the nitrous acid itself, only indirectly, therefore, sulphonating the nitrite of a metal or of ammonium. One third of the pyrosulphite should, accordingly, be replaceable by some other acid, and so it proves to be (section III. d). It is not new to formulate the sulphonation of IINO<sub>2</sub>, and to speak of 'nitrous acid' as the reacting substance, for (passing over Fremy) Raschig has already done so. But, whereas we would be understood to confine the activity to nitrous acid itself, or its acidic equivalents (sect. III. d), such was not the thought of Raschig, who only wrote H as a general symbol, and 'acid' as a general term, while representing metal nitrites as active by generating alkali hydroxide.

Nor is it new to learn that nitrous acid can be sulphonated. By treating a dissolved sulphite with nitrous acid (nitrous fumes) Fremy did succeed in obtaining sulphazotised salts, but the difficulty of moderating the flow of the gas, and the presence in it of nitrogen peroxide and nitric acid made the operation so inconvenient, he said, that he did not use it in preparing any of the salts he examined, and gave no further attention to it. We have taken up the matter, since untouched and unmentioned, where Fremy left it half a century ago. Our work has been very simple but very effective, and has consisted in subjecting a

solution of pyrosulphite (and of normal sulphite, but of that we treat in sect. III. b.) to nitrous fumes which act as nitrous anhydride when of the right composition. The gases were found to be fully absorbed by a concentrated solution of potassium pyrosulphite kept cold in a flask immersed in ice and brine and well agitated. Soon an abundant precipitation began of hydroximidosulphate mixed with a little nitrilosulphate. While still much pyrosulphite remained, the process was stopped and the mother-liquor at once drained off. In this way we had great success in getting much hydroximidosulphate and only a little nitrilosulphate, notwithstanding the presence all along of so much pyrosulphite; for, as was pointed out by us long ago, in sufficiently cold solutions sulphonation to nitrilosulphate hardly occurs.

The next five sections (III. b, c, d, e, f) treat of various mixtures which, from the acid constitution of one of the components, behave like that of nitrite and pyrosulphite, that is, as if each contained pyrosulphite and nitrous acid.

## III.—b. Normal Sulphite also all active upon Nitrous Acid.

Replacing the pyrosulphite used in the last experiment by the normal sulphite, it was found that again but in this case gradually, hydroximidosulphate precipitated, as well as very little nitrilosulphate. But here potassium nitrite proved to be another product, which by gradually replacing the potassium sulphite in the solution allowed the process to be carried very far towards completion. The reaction is expressed by the equation—

3HNO<sub>2</sub>+2K<sub>2</sub>SO<sub>3</sub>=2KNO<sub>2</sub>+OH<sub>2</sub>+K<sub>2</sub>HNS<sub>2</sub>O<sub>7</sub>, from which it is seen that only one-third of the nitrous acid becomes sulphonated, the rest being used up simply as an acid.

This interaction is what, we believe, Raschig must inadvertently have got, when seeking to prepare Pelonze's salt (hyponitrososulphate) by the use of nitric oxide. The conditions are favourable to the production of the nitrito-hydroximidosulphate (this vol., p. 222).

## III.—c. Action of Sulphur Dioxide upon Normal Sulphite and Nitrite.

It has been shown in this paper (sect. I. b) that the hydroximidosulphate which, from the first, accompanies the normal sulphite as joint product of the action of sulphur dioxide upon alkali nitrite and hydroxide, keeps steadily to small proportions to the sulphite until nearly all the hydroxide has been saturated. After that point is passed and when, therefore, sulphur dioxide is meeting a mixture of nitrite and normal sulphite, examination of the solution, by the method already described, shows that, along with a greater production of hydroximidosulphate than before, there is pyrosulphite produced in no insignificant quantity. This remarkable growth in the quantity of pyrosulphite, considered along with the fact (sect. II. a) that it is itself active upon nitrite proves that much of the sulphur dioxide goes altogether to the normal sulphite. Only after the greater part of this salt has been acidified to pyrosulphite is the sulphur dioxide active in sulphonating the nitrite, which it then does by combining with it in conjunction with the pyrosulphite, thus:—

2KNO<sub>2</sub>+K<sub>2</sub>S<sub>2</sub>O<sub>5</sub>+2SO<sub>2</sub>+OH<sub>2</sub>=2K<sub>2</sub>HNS<sub>2</sub>O<sub>5</sub>, the hydroximidosulphate being produced in this way with much greater facility than by the pyrosulphite alone because of its production not being accompanied here by the regeneration of normal sulphite with its inhibitory effect upon sulphonation (sect. II. a). In this change it still holds true that it is nitrous acid itself which is sulphonated, the potassium leaving the nitrite to enter the sulphonate radical, and being replaced by hydrogen.

Claus held that there could be no difference between the effect of submitting a nitrite to the action of a sulphite and that of mixing it with a solution of hydroxide and then treating it with sulphur dioxide. The contents of this section and section II. a show that essential difference exists between the courses and results of the two procedures.

## III.—d. Action of Carbon Dioxide and of an Acid Carbonate upon Normal Sulphite and Nitrite.

As would be expected, the gradual addition of one of the stronger acids to a solution of normal sulphite and nitrite leads to the formation of sulphazotised salts. But even carbon dioxide and the acid carbonates of the alkalis are effective in exciting action in a solution of these salts. Concerning the activity of carbon dioxide there is nothing to add to what was published in our first paper (J. Ch. Soc., 1887, 51, 661), that the gas is very slowly absorbed by the mixed salts in solution though not by either salt alone and at the mean temperature, and that sulphazotised salts are then produced. Normal carbonates of the alkalis are inactive.

It is known that nitrites are not decomposed by carbon dioxide, and also that alkali carbonates are decomposed by pyrosulphites as freely at the mean temperature as by sulphur dioxide itself. Accordingly, we have found that potassium or sodium acid-carbonate dissolved along with excess of normal

potassium or sodium sulphite gives off carbon dioxide to a current of decarbonated air much to the same extent as when dissolved alone in water. But sodium acid-carbonate may be added to an ice-cold solution of sodium pyrosulphite, containing also much normal sulphite, and be only very gradually decomposed with effervescence. Indeed, an ice-cold concentrated solution of normal sodium sulphite will deposit some acid-carbonate when charged with carbon dioxide.

It is, therefore, not surprising that sodium or potassium acid-carbonate has a very marked action upon mixed normal sulphite and nitrite. When the three salts are left together in solution in a closed vessel for a day or two, much sulphazotised salt is formed, so that after carbonate and excess of sulphite have been precipitated by baryta and barium chloride in presence of ammonium chloride, the filtrate from the precipitate when boiled with acid gives much barium sulphate and reduces cupric hydroxide freely. The interaction of the salts may be expressed by the equation—KNO<sub>2</sub>+2K<sub>2</sub>SO<sub>3</sub>+3KHCO<sub>3</sub>=K<sub>2</sub>HNS<sub>2</sub>O<sub>7</sub>+3K<sub>2</sub>CO<sub>3</sub>+OH<sub>2</sub>, but since the two-thirds normal hydroximidosulphate is to a small extent converted by normal carbonate into a more nearly normal salt and acid-carbonate (this Journal, 7, 32), the change expressed by the above equation cannot proceed to completion.

## III.—e. Action of Sulphur Dioxide upon Normal Carbonate and Nitrite.

When sulphur dioxide is added to two mols, nitrite and one mol, normal carbonate until the solution becomes acid to lacmoid paper, the only products are hydroximidosulphate and carbon dioxide. This was long ago pointed out by us, and also that sulphite and acid carbonate are intermediate products, the latter of which separates for a time from concentrated solutions. We have made further experiments to ascertain the effect of the first portions of the sulphur dioxide in producing hydroximidosulphate, which, where alkali hydroxide is used, we have shown to be insignificant.

These experiments were carried out in the same way as those for testing the effect when sodium hydroxide is employed (I. b) but with the modification of making two pipettings each time instead of one, and of weighing both instead of merely measuring them, then in the one we determined the sodium, as sulphate and used the result for calculating what fraction of the original solution the other quantity was in which we determined sulphite and sulphonates. We thus made ourselves independent of the change of volume during the reaction caused by loss of carbon dioxide and gain of sulphur dioxide. We found in this way, admitting of no refined accuracy, that at a later sampling the solution contained at most, as much as 3½ per cent. less sodium than at an earlier sampling, a difference however hardly large enough to need attention.

The flask for receiving the portion for the sodium determination was previously weighed empty but that for the other portion was weighed containing some concentrated solution of sodium hydroxide, placed there to arrest all action in the pipetteful dropped into it. In the first portion could be seen, by its changes on standing, how necessary the sodium hydroxide was for fixing the composition of the solution at the time it was sampled; sometimes acid carbonate was deposited by it, sometimes hardly at all: sometimes the precipitated acid carbonate slowly disappeared sometimes not. The solution used contained

1 part of sodium nitrite in 4.64 parts of water, besides the calculated quantity of anhydrous sodium carbonate.

The results of the experiments showed that hydroximidosulphate was largely produced from the beginning, in proportion to the sulphite also formed. Thus, in one experiment, when 25 per cent. of the sulphur dioxide required for complete sulphonation had been passed in, 55.3 per cent. of it had become sulphonate, the rest (44.7 per cent.) sulphite. When 53.6 per cent. of the sulphur dioxide required had been used, 74.9 per cent. of it had become sulphonate and 25.1 per cent. sulphite. In another closely comparable experiment, when 33.7 per cent. sulphur dioxide of that required had been absorbed, 62.7 per cent. of it had become sulphonate and the rest sulphite; when 44.4 per cent. of the whole had been used, 72.75 per cent. of it had become sulphonate; and when 62.2 per cent. of the whole had been used, 81.5 per cent. of it had become sulphonate. That is to say, as for the last statement, when 20.2 grams of sodium nitrite (with carbonate) had received 37.5 grams sulphur dioxide, 23.3 grams of this had become sulphonate and 14.2 grams had become sulphite.

Uniform results are here, however, as when hydroxide is started with, only obtained by uniform working, of which the following experiment is a good example. A solution of sodium nitrite and carbonate was divided approximately into one-fifths and four-fifths, and both portions were treated, as nearly as could be, alike, their unequal quantities making the only difference. The smaller portion when it had received 20 per cent, of the full amount of sulphur dioxide was found to contain 61.8 per cent, of it in form of sulphonate, 38.2 per cent, of it as sulphite. The larger portion, having received 25 per cent, of the amount necessary for

its full sulphonation, was found to have only 53.3 per cent. of it as sulphonate and 44.7 per cent. of it as sulphite, as already given; had we stopped here at 20 per cent. sulphur dioxide, as we did with the smaller portion, the difference would have been more striking still. The difference observed was due to the smaller portion having, in relation to its quantity, received sulphur dioxide four times more rapidly than the larger portion had, the stream of sulphur dioxide having been steady and closely alike in the two cases. The result was that local saturation was less checked by the agitation of the flask in this case than when the much larger portion of solution was under treatment.

The lack of uniformity in the results here described, does not affect in the least the evidence they afford that the sulphonation of nitrite in presence of carbonate differs greatly in its course from that it runs in presence of alkali hydroxide.

Respecting the formation and destruction of sulphite in the process, this salt was observed to be produced rapidly until in quantity it had become equivalent to about one-eighth of the sulphur dioxide needed for sulphonation of all the nitrite. Then, for a time, its quantity remains nearly steady, all sulphur dioxide entering the solution during that time becoming sulphonate. Finally, it steadily lessens in quantity as more sulphur dioxide is added, and disappears just at the end of the sulphonation. The more rapidly the sulphur dioxide is blown in at first, the less of it becomes sulphite, and the more sulphonates, as already stated above.

One other striking thing observed in these experiments was the great variability of the point at which acid carbonate first precipitated, as well as the variability of its quantity. With quick working acid carbonate precipitated much earlier and in much larger quantity than in slow working, proportionately, that is, to the fraction the solution had received of the quantity of sulphur dioxide needed for complete sulphonation of the nitrite. Thus, while, with quick working, acid carbonate separated in abundance when 20 per cent. of all sulphur dioxide had been absorbed, it only precipitated, and then much less copiously, when 44 per cent. of all sulphur dioxide had been supplied relatively more slowly to the solution. In another experiment it showed itself only when 53 per cent. of the sulphur dioxide had been added. The main condition, therefore, for early precipitation of acid carbonate is rapid addition of the sulphur dioxide at first,—the same condition as favours growth of sulphonates at the expense of sulphite.

Now for the discussion of the results. It becomes highly probable from a consideration of these results, together with what we know of the several substances concerned, that the first action or tendency to act of sulphur dioxide when it enters the solution is to convert carbonate into normal sulphite and acid carbonate, and to leave the nitrite untouched, and that this action remains prominent so long as much normal carbonate is undecomposed. Though this cannot be shown experimentally, it is certain that this action does take place, for its products present themselves freely, products which could not be derived from the sulphonation of the nitrite. Both normal sulphite and acid carbonate are active along with sulphur dioxide in sulphonating nitrite.

In accordance with what is stated in III. d. the normal sulphite and acid carbonate together slowly disappear of themselves from the solution when addition of more sulphur dioxide

is stopped, owing to sulphonation of the nitrite and reconversion of acid carbonate to normal carbonate—

NaNO<sub>2</sub>+2Na<sub>2</sub>SO<sub>3</sub>+3NaHCO<sub>3</sub>=Na<sub>2</sub>HNS<sub>2</sub>O<sub>7</sub>+3Na<sub>2</sub>CO<sub>3</sub>+H<sub>2</sub>O. such a mode of sulphonation will therefore be also in operation when the entrance of more sulphur dioxide has not been arrested, but it is very slow in presence of normal carbonate and may be disregarded as a factor in the process of sulphonating when sulphur dioxide is also at work. Here we would insert that only to simplify discussion do we speak of normal sulphite and carbon dioxide, or even acid carbonate, being together unchanged; these substances, as previously stated, act on each other to a large extent in ice-cold solutions, and in our work we met with precipitated acid-carbonate at times when it could only be there in consequence of carbonic acid withholding sodium from pyrosulphite.

That in the earlier stages of the process, when much carbonate is present, the normal sulphite plays a very small part in the sulphonation not only follows from the observation of its rapid increase in quantity at first but is also shown by its then nearly constant quantity for a long time though sulphur dioxide is still entering the solution and forming sulphonates. Only later, as the carbonate gets consumed, does the sulphite become an important factor in the sulphonation by freely becoming pyrosulphite, for then its quantity rapidly falls.

The part played by sulphite in the early stages being thus insignificant, we have to seek in the carbonates the source of the early considerable sulphonation of the nitrite. It would be unreasonable to assume, with acid carbonate present, that the normal carbonate takes part in sulphonation; equally so to assume that it remains inactive to sulphur dioxide. We are therefore compelled to recognise that sulphonation goes on only after conver-

tion of all carbonate *locally* present to acid carbonate and sulphite has been effected. Then the reaction that ensues is—  $NaNO_2 + NaHCO_3 + 2SO_2 = Na_2HNS_2O_7 + CO_2$ 

When all normal carbonate in the solution has been acidified by the carbon dioxide, the sulphite becomes as active as the acid carbonate and neither salt gets consumed before the other.

While it seems certain that first the sulphur dioxide converts the normal carbonate into normal sulphite and acid carbonate, and only then produces hydroximidosulphate by acting on the nitrite along with acid carbonate in the earlier stages and on both this and normal sulphite collaterally in the later stages, the experimental results show that local saturation must take place largely where the sulphur dioxide enters the solution, since so much sulphonate is produced along with the sulphite. In consequence of the activity of acid carbonate, local saturation becomes twice as difficult to prevent as when hydroxide is used in place of carbonate.

If in order to impede local saturation we slacken the rate of passage of the sulphur dioxide into the solution, we meet with a good amount of success. Thus, it was shown by the results of experiments already given, that the slower rate gave proportionately less sulphonate and more sulphite. But the effect of slowness in passing in the gas has its limit, in consequence of the continuous though slow interaction which takes place between nitrite, normal sulphite, and acid carbonate whereby sulphite disappears to give place to sulphonate. It follows that too slow as well as too rapid an addition of sulphur dioxide is unfavourable to the accumulation of sulphite, rather than of sulphonate, in the solution, and that a medium rate of supply is best for raising the proportion of sulphite.

There remains to be explained the great variability in the commencement of precipitation of the acid carbonate. This takes place the sooner the faster the sulphur dioxide is blown into the solution. When it occurs in the earlier stages of the process, it is, therefore, accompanied by greater predominance than usual of production of hydroximidosulphate over production of sulphite. It does not however depend upon this, for while sulphur dioxide liberates a molecule of earbon dioxide in changing carbonate into sulphite, four mols. of it are needed to liberate one mol. of carbon dioxide in changing carbonate and nitrite into hydroximidosulphate.

An explanation is suggested by a consideration of the fact that when working the process at a moderate rate, the first crystallisation of acid carbonate takes place long after the point at which the solution must contain the maximum of the salt, at least potentially, the point, that is, when half the carbonate has become either sulphonate or sulphite. When it does occur the quantity of it in solution has become much less. Only where crystallisation is started early by a very rapid addition of sulphur dioxide, does the acid carbonate continue to separate out in much such quantity as it could do at the stage of the process reached. The cause in one word is supersaturation. The acid carbonate, it would seem, is slow to begin to precipitate from the solution while that is not charged with carbon dioxide. At a medium rate of working this only happens in the later stages, any normal carbonate and even much normal sulphite present keeping down the quantity of carbon dioxide, but by a rapid rate of working local saturation occurs and the acidified portion of the solution then crystallises. Once crystallisation has been started, it proceeds unchecked. In slower working when crystallisation only begins

late in the process, the amount of salt separating is small, and generally depends then for its existence upon its power to resist the action of acid sulphite in ice-cold solutions. The solution when, potentially at least, it is richest in acid carbonate, was found by us to crystallise soon, if left to stand in closed vessel, although sulphonation which is destructive of acid carbonate was slowly going on in it.

## III.—f. Primary Action of Sulphur Dioxide upon a Nitrite.

Solution of sulphur dioxide added to that of potassium or sodium nitrite produces a sulphate and either nitric or nitrous oxide, according as one or other of the interacting substances is in excess. That is the ordinary well-known result, but there are two ways of limiting the extent of the action so as to get either hydroximidosulphate and nitrous acid or the undoubted products of their transformation. By these ways, the interaction of sulphur dioxide and a nitrite is shown to be—

$$2KNO_2 + 2SO_2 + OH_2 = K_2HNS_2O_7 + HNO_2$$
.

The more important way to thus limit the action is by an experiment first tried by Claus (Ber., 1871, 4, 508; see preceding paper) which consists in adding an alcoholic solution of sulphur dioxide to excess of potassium nitrite in strong aqueous solution. For this experiment gives, as we have ascertained, potassium nitrito-hydroximidosulphate which precipitates and ethyl nitrite which boils off by the heat of the reaction:—

$$3KNO_2 + 2SO_2 + C_2H_6O = KNO_2, K_2HNS_2O_7 + C_2H_5NO_2.$$

By becoming ethyl nitrite the nitrous acid is rendered inactive on the hydroximidosulphate, which is thus saved from oxidation.

The other way of tracing the earlier action of sulphur dioxide

upon a nitrite was found out by Raschig, when trying to prove another point (sect. IV. a.). He added the nitrite to excess of sulphur dioxide, both being in very dilute and well cooled solution, evaporated down and neutralised the solution with chalk, and again evaporated the filtered solution. After much potassium sulphate had crystallised out, potassium amidosulphate was finally obtained, as proof that hydroximidosulphate had been formed at an earlier stage. Our own experiments have yielded us an earlier product of the degradation of this compound.

At the time when Raschig published his observation, we published (J. Ch. Soc., 1887, 51, 659) one of ours, that silver nitrite and mercurous nitrite, when decomposed by sulphur dioxide solution, yield a substance answering to the copper test for hydroxylamine. This we now know to be hydroxylamine sulphuric acid, but at the time we took it to be hydroxylamine itself. We have also found that, after adding a dilute solution of sodium nitrite to excess of a cooled solution of sulphur dioxide and then blowing out of the solution the residual sulphur dioxide by a current of air, enough hydroxylamidosulphate (hydrolysed hydroximidosulphate) is present to be easily identified by the copper test for it. A hydroxylamidosulphate is distinguishable from hydroxylamine in applying this test by finding that the mother-liquor of the cuprous oxide (which need not be filtered off) gives sulphurous acid when acidified (this Journal III, 225).

Though less successful than Claus's experiment, Raschig's method is serviceable for showing that the alcohol used in that, plays only a secondary part. While excess of nitrite is successfully used in that experiment, the sulphur dioxide must be in excess in Raschig's method. To understand this, it has only to

be remembered, firstly, that nitrous acid would oxidise hydroximidosulphate at once, and secondly that sulphurous acid sulphonates the hydroximidosulphate slowly enough to allow a little of it being secured in a hydrolysed state.

#### IV.—a. Sulphonation of Nitrous Acid by Sulphurous Acid.

Fremy believed that certain of his sulphazotised salts are formed in the first action of sulphurous acid upon nitrous acid. From this belief Claus strongly dissented, holding that the presence of a base (as salt) was essential to the production of these acids. Raschig considered that his experiment of treating potassium nitrite with sulphur dioxide in excess (sect. III. f.) proved the correctness of Fremy's belief; but that cannot be admitted since potassium is present in this experiment playing the part of base. It is, however, quite practicable to establish Fremy's belief and that no base whatever is necessary to bring about the formation of sulphazotised acids.

When a solution of sulphur dioxide, better ice-cold, is treated with a relatively small quantity of nitrous fumes passed on to its surface while it is being well agitated in a flask, and is then deprived of remaining sulphur dioxide by a rapid current of air, or even by quick boiling, it will give a good reaction for hydroxyamidosulphuric acid with the copper test. A little deviation in the composition of the nitrous gas from that of nitrous anhydride is not of importance. If the object is only to get amidosulphuric acid, the solution of sulphur dioxide is left to stand for a day after it has received the nitrous acid without expelling what is left of the sulphur dioxide. If it is then evaporated on the water-bath and further concentrated in the

vacuum-desiceator, the amidosulphuric acid will crystallise out from the sulphuric acid with which it is accompanied (this Journal, 9, 230). We have purified the acid by recrystallisation, and have hydrolysed it at 150°, by means of hydrochloric acid, into acid ammonium sulphate; we have also completely volatilised the acid by heat thus proving the absence of base accidentally derived.

Nitrosyl sulphate dropped into much excess of cooled solution of sulphur dioxide also yields the hydroxyamidosulphate reaction with copper sulphate and potassium hydroxide.

#### IV.—b. Influence of the Base of the Nitrite or Sulphite.

Although Fremy held that sulphurous and nitrous acids combine together, he did not believe that the resulting sulphazotised acids could be obtained in this way, because of their inability to exist in absence of a base. Moreover, he considered that a strong base is influential in bringing about the formation of these acids, even though he had had no success with such a base as sodium. The only hydroximidosulphates he could prepare, indeed, were those of potassium, but from ammonium nitrite he got the nitrilosulphate, and also obtained evidence that calcuim, strontium, and barium nitrites are convertible into amidated sulphates.

We have just shown (sect. IV. a.) that the interaction of sulphurous and nitrons acids does not require the presence of any base at all for the actual production of sulphazotised acids, although such presence is essential to preserve unchanged the first product of the interaction. To serve this purpose some bases will doubtless be inferior to others, and those which do

not freely form soluble pyrosulphites are difficult to work with. Otherwise, the nature of the base seems to be a matter of indifference. Since the time of our early publications on the subject, we have extended our experiments to several other nitrites than those of sodium, mercurosum, and silver, with the results we now record.

Ammonium salts.—Ammonium nitrite solution was prepared by triturating silver nitrite with its equivalent of ammonium chloride dissolved in about five times its weight of water, and filtering off silver chloride over the pump. To this solution, after it had been cooled in ice, was added a little less than its equivalent of ammonia-water which had just before been converted to sulphite by passing sulphur dioxide into it. More sulphur dioxide was then passed into the mixture until it reddened lacmoid-paper. In this way the ammonium nitrite was almost all sulphonated, without any evolution of gas having occurred till just at the last, when slight nitrous fumes appeared. Some of the solution was hydrolysed and tested then with copper sulphate and potassium hydroxide; it was thus shown to have contained abundance of ammonium hydroximidosulphate. Another portion of the solution not hydrolysed gave a large precipitation of dipotassium hydroximidosulphate on addition of potassium chloride.

Barium salts.—Some barium hydroxide was converted into sulphite by putting it in water and passing in sulphur dioxide; the barium sulphite was then, for the most part, brought into solution by passing in more sulphur dioxide. The product was added gradually to a solution of a little more than its equivalent of barium nitrite, which had been purchased of excellent quality.

Having neglected to cool our solutions we had reason to fear that our experiment was a failure; for along with very much precipitation there was a somewhat large evolution of nitrous gases. But for our purpose we had been amply successful. The solution was only faintly acid to litmus and remained so for hours. Both it and the precipitate contained large quantities of barium hydroximidosulphate. The precipitate also contained sulphite and sulphate, the latter being the complement to the nitrous fumes produced. The hydroximidosulphate was extracted from the precipitate by a solution of ammonium chloride.

Calcium salts.—A solution of calcium nitrate, free from magnesium, sodium, potassium, and other ordiary impurities, was heated with well-washed spongy lead until nitrogen oxides and ammonia began to form. The filtered, very alkaline, solution was freed from lead by hydrogen sulphide not used in excess, Calcium hydroxide was then removed by carbon dioxide. (it was interesting to find that, contrary to assertion, carbon dioxide cannot be used to precipitate lead in presence of calcium salt, since calcium precipitates before lead.) A solution of calcium sulphite in sulphurous acid was prepared just before use, in the same way as the barium salt had been, except that carefully prepared calcium carbonate took the place of barium hydroxide. With the calcium nitrite somewhat in excess of the calcium sulphite, the solution of the latter was gradually poured into the former, both solutions having ice floating in them at the time. No gas was given off and only a moderate quantity of precipitate was formed, which consisted of sulphite. The filtrate was neutral and contained the full quantity of hydroximidosulphate expected.

Zine salts.—Zine nitrite solution was prepared by precipita-

ting zine sulphate with barium nitrite and filtering. Zine sulphite in solution in sulphurous acid was made from zine oxide in water and sulphur dioxide. The two solutions, suitably proportioned and with ice floating in them were mixed. No gas came off, zine sulphite precipitated, and the solution proved to contain zine hydroximidosulphate present in it in large quantity.

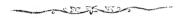
Mercurous salts and silver salts.—Experiments, already referred to in sect. III. f. of this paper, sufficiently establish that mercurous and silver nitrites are readily sulphonated. It is now evident that the sulphonation of nitrites is a general reaction, essentially independent of the nature of the base, which only effects the preservation of the products. It is not the salts which are sulphonated but nitrous acid itself.

## V.— What Nitrous Acid becomes when Sulphonated.

In the paper preceding this it has been established that neither the abundant experimental work of other chemists and ourselves nor theoretical considerations afford any support to the view that the double sulphonation of nitrous acid into a hydroximidosulphate occurs in two stages, or that a monosulphonated nitrous acid, ON·SO<sub>3</sub>H or (HO)<sub>2</sub>N·SO<sub>3</sub>H, must be the first product of its change. In the present communication it is shown that the acidity necessary for the sulphonation of a nitrite points clearly to the fact that it is in every case the acid itself, and not its salts, which is directly sulphonated. We are, therefore, in the position to affirm that the fundamental action in the formation of all Fremy's sulphazotised salts is the interaction between actual nitrous acid and a pyrosulphite, in which they unite always to form the one substance, the two-thirds normal hydrox-

310 divers & haga: interaction betw. sulphites & nitrites.

imidosulphate corresponding to the pyrosulphite acting—HONO+(SO<sub>2</sub>K)·SO<sub>3</sub>K=HON(SO<sub>3</sub>K)<sub>2</sub>. The origin of all the other salts out of this salt has been traced, partly by others and partly by ourselves, and need not be gone over again here.



# Contributions to the Morphology of Cyclostomata.

II.—The Development of Pronephros and Segmental Duct in Petromyzon.<sup>1</sup>

By

#### S. Hatta,

Professor in the College of Peers, Tōkvō.

With Plates XVII-XXI.

The following pages contain the second of a series of studies on the later stages in the development of *Petromyzon*, the first having already been published some time since in this journal ('97, vol. x, pp. 225-237).

Our knowledge of the earliest development of the excretory organs in the lampreys is still somewhat incomplete. This circumstance is, I believe, mainly due to the want of recent investigations upon the subject. Since the appearance of the works by Müller ('75), Scott ('82), Shipley ('87), Goette ('88), Kupffer ('90), and others, ten years or more have

<sup>1)</sup> It was my intention to publish this paper shortly after the appearance of my preliminary notice in 1897, (Annot. zool. Jap., vol. I. pp. 137-140) but various unavoidable elecumstances have combined to cause the delay. Meanwhile I have had opportunities of renewing my study on various points and the results here given are different from those of the preliminary paper in several important respects.

elapsed, and, so far as I am aware, no important fact has been added during the interval by any renewed researches. I need not, therefore, apologise for the publication of the present paper which embodies the results of my study on the subject during the last few years.

The investigation of a longitudinally stretched, or a metamerically arranged, organ-system such as the neural canal, the chorda, the pronephros, &c., is rendered peculiarly difficult in Petromyzon by the fact that the longitudinal axis of the embryo in early stages describes a semi-circle. Some sections in a series of cross-sections of such an embryo are therefore unavoidably cut in planes which meet the longitudinal axis of the embryo in variable degrees of inclination; consequently a structure stretching in the direction of this axis is cut through obliquely, as, for instance, the neural cord shown in figs. 2 and 3, Pl. xvii. The vertical dimension of the cord is not in reality as long as is represented in these figures. To gather accurate notions of the form, the position, &c., of a given structure, therefore, it is necessary to compare series of sections of two or more embryos of as nearly the same age as possible. Further, the difficulty of observation is greatly increased by the crowd of yolk-granules in cells, especially by their reaction against staining fluids. Certain fluids such as haematoxylin, borax-carmine, &c., either stain diffusely all the parts, or act on the granules more intensely than on the other contents of cells, so that we can not discriminate different kinds of tissues. This difficulty was, however, obviated by employing picro-carmine. The embryos were stained in toto in this fluid, decolorized to the proper degree in acid-alcohol, and then washed in 90% alcohol. In the sections of specimens thus prepared, the histological structures are distinguishable very clearly, being almost entirely discolored in all parts except nuclei which are stained intensely.

I wish here to express my warmest thanks to my former teachers, Prof. Mitsukuri and Prof. IJIMA, for much invaluable advice and for their kindness in looking through the manuscripts and the proof-sheets of this paper.

To avoid confusion the present paper will be divided into two sections, the first of which will contain mere descriptions, while in the second will be given a historical review and conclusions.

### I. Descriptive.

### A.—The Pronephros.

The youngest stage of the embryo which I have to deal with in the present investigation, is only a little advanced beyond an ellipsoidal gastrula: it is intermediate between Stage I and II of the list given in my first contribution above referred to (fig. 1, A and B, of that article). The head-fold forms a pointed protuberance at one pole of the ellipsoid, while the blastopore is plainly visible at the opposite pole. The prominent neural ridge extends longitudinally from the anterior end of the head-protuberance to the dorsal lip of the blastopore.

For the general relation of the germinal layers at this stage, I refer to fig. 1 which is drawn from the same series as the section represented in fig. 18 of my former work.<sup>1)</sup> It represents a transverse section though the dorsal region of an embryo in the stage above described. As seen in this figure, the solid

<sup>1)</sup> S. Hatta, On the form, of the germ, lay, in Petr.; this Journal, vol. V, 1891.

314 s. hatta:

neural cord is interposed in the median line between the right and left halves of the mesoblast, underneath the epiblast composed by of a single row of columnar cells. The epiblast is always limited with a sharp contour against the mesoblast. The latter consists, on each side, of a dorsal (d.), a ventral (v.) and a median (m.) row of cells. The dorsal row represents the parietal, and the median and ventral rows together form the visceral, layer. Both the visceral and parietal layers of the mesoblast show, in the proximal portion, a regular arrangement of a high columnar epithelium, while distally this arrangement is more or less disturbed.

The above structures are localized in a small portion on the dorsal aspect of the ovoid embryo, the remaining larger part being taken up by yolk-cells compactly loaded with yolk-granules.

In this early stage, we can detect, therefore, neither in the epiblast nor in the mesoblast any structure whatever which is to be regarded as the rudiment of the pronephros.

#### Period 1.

In the next following stage, which corresponds approximately to the early part of Stage II (loc. cit., fig. 1, B), certain alterations are met with in the mesoblast. The most important of these is its metameric segmentation. This process first begins at the neck<sup>1)</sup> and proceeds both forwards and backwards. At the present stage, there are found 16 or more mesoblastic somites.<sup>2)</sup> At about the time when this process has extended to the anteriormost section of the mesoblast a second change arises in the mesoblast, viz. the first appearance of the pronephros.

<sup>1)</sup> The term nerk is used for the sake of convenience to designate the slender region where the head-fold passes over into the hind globular part.

<sup>2)</sup> The exact number of the somites can not be reckoned, for the metameres become indistinct posteriorly.

Fig. 2 represents a section through the middle of the fifth somite<sup>1)</sup> of the embryo mentioned above. The general features of the germinal layers and of other primitive organs are essentially the same as before. The epiblast (ep.) is a single row of columnar cells and is sharply bounded from the structures beneath it; the neural cord (n.) remains still solid.<sup>2)</sup> In the mesoblast, however, two portions are distinguishable: the proximal portion composed of high columnar cells (mt, V) and (a.p.n. 2) which undergoes metameric segmentation, and the distal portion consisting of a loose group of somewhat irregularly shaped cells (lm.) which remains unsegmented and constitutes the lateral plate. is noteworthy that the former takes up the largest portion of the mesoblast, while the latter is represented by a small portion; these two portions represent respectively the parts of the same name in the mesoblast of Amphioxus. However, between them there exists no distinct limit in the lamprey; the one passes gradually over into the other. Although the visceral layer shows no sign of constriction, the parietal layer is notched at about the middle of the proximal segmented portion (x). The parietal layer distal to this notch is composed of a regular cylindrical epithelium (a.pn. 2), which is slightly arched against the epiblast, so as to cause an indentation in the latter, while the visceral layer of the corresponding portion consists of a more or less disturbed row of high columnar cells. As the subsequent history teaches, the proximal half of this extent (mt.1) represents the myotome<sup>3)</sup>

<sup>1)</sup> The somites are reckoned from the anterior end. The first, i.e. the foremost lies immediately behind the auditory vescicle when the vescicle comes into view.

<sup>2)</sup> The vertical diameter of the neural cord in sign, 2 and 3 is shown greater than it really is, the sections passing obliquely owing to the bending of the longitudinal axis of the embryo, as noted in the introduction (p. 312).

<sup>3)</sup> This term here means the Sciero-myotom of German authors.

and the distal portion (a.pn.2) constitutes the Anlage of the pronephros—the name I assigned to the same in my preliminary paper ('97). To avoid tiresome reiteration, I shall often speak of them in the following pages simply as the "Anlage" and when it is necessary to refer to special ones, as the Anlage first, the Anlage second, etc. in the order of their position in the series of mesoblastic somites, beginning from the anterior end.

In the somite next following, i.e. the sixth (fig. 5), the mesoblast shows almost the same condition as that already described; but in the somite preceding the fifth, i.e. in the fourth, the Anlage of the pronephros is a little more advanced in development (fig. 3). On the left side of fig. 3, the fourth myotome is sliced only at its hind wall (mt.IV), while, on the right, it is cut through in the middle (mt.II). On the right half of the section, no marked progress is visible in the mesoblast except the separation of the myotome, which shows a pentagonal outline (mt.IV) and consists of high cylindrical cells from the lateral plate formed of a loose mass of cells (lm.). In the left half, however, the state of things is quite otherwise: the Anlage of the pronephros (a.p.a. 1) together with the corresponding visceral layer is entirely constricted off from the myotome (mt.11), although it is still connected with the lateral plate (lm.). The cells composing the Anlage are compactly set together and arranged more or less in a radial manner; the Anlage itself is rounded off at the proximal end. The lateral plate, on the other hand, still consists of loosely grouped cells of variable shape.

The Anlage is thus always (before and after its separation from the myotome) histologically very distinct from the lateral plate; one might therefore often be misled to suppose that there is no organic connection between these two structures.

Fig. 4 represents a section passing between the two somites above mentioned (the fourth and fifth) and is much magnified (Zeiss,  $E \times 2$ ) to illustrate the finer structure of this portion. The structural cells are all loaded with an enormous quantity of ovoid corpuscles or yolk-granules. The epiblast (ep.) consists of a single row of cubical cells and shows a sharp limit against the structures inside it. The irregularly polygonal mass of cells (ml. T) is the anterior wall of the fifth myotome. Two rows of variously shaped cells (lm.) constitute the lateral plate which is histologically quite like that in the somitic portion, being composed of irregularly quadratic cells and tapering towards the distal (ventral) extremity (compare with the lateral plate, However, in the proximal portion, lm., in figs. 2 and 3). where the Anlage of the pronephros consisting of a regular row of tall columnar cells would be found in the somitic portion, we see here a group (x) of a few cells of faint appearance, forming the proximal edge of the lateral plate. By a comparative study of two or more series of sections, it is easily demonstrated that these cells are a piece of the somite lying in front and have nothing to do with the Anlagen. To elucidate this point still further, I have drawn fig. 7 which represents a section through the intersomitic plane between the sixth (fig. 5) and the seventh somite (fig. 6). In this part the Anlage of the pronephros is developed still more weakly, and the mesoblast remains in a more primitive state. In the proximal edge of the lateral plate (x), no special structure is detected, but the edge fades away without a distinct limit. By comparison with Fig. 4, we can not find any marked difference; thus, here likewise, there is no cellular connection between the Anlagen in the two succeeding somites.

From the fifth somite backwards for 9 or 10 somites, the

318 s. hatta:

mesoblast presents almost the same feature of the Anlage as in the fifth somite mentioned above. Fig. 5 represents a section through the sixth somite, next behind the fifth; when compared with fig. 2 no marked difference is detected in regard to the structure of the mesoblast. But in some segments the development of the Anlage is somewhat weaker than in others, as seen in fig. 6, which shows a section through the seventh somite; while in a segment posterior to this somite, we find the Anlage as much pronounced as in the sixth somite. However, generally speaking, the Anlage of the pronephros in an anterior somite develops further than that in a posterior. It must be remembered that the somite in which the Anlage has already become expressed does not pass over suddenly into the somite in which no trace of it is to be seen; but its development gradually grows less and less distinct from the anterior to the posterior part, until finally no trace of it is perceived.

In the present stage, therefore, the Anlage of the pronephros is detected in more than 4 somites but is completely separated from the myotome only in one segment, viz. the fourth somite<sup>1)</sup>, and it has no genetic connection either with the Anlage in the next following somite or with the epiblast: and it must be noticed that we find the foremost Anlage not exactly beneath the fourth myotome, but always underneath its hind border.

Figs. 8-17 represent sections through a still older embryo of this stage, having about 20 somites. The epiblast (ep.), the neural cord (n.), and the chorda (ch.) are essentially the same as before. Being cut through somewhat obliquely, the myotomes

<sup>1)</sup> Such a case is very rare. In most preimens examined, the Anlage separated from the myotome is found in many segments, so that we can hardly decide in which segment the separation takes place first.

on the two sides do not exactly correspond. On the right side of fig. 8, the hind border of the fourth myotome (mt.II') is cut through; the Anlage of the pronephros (a.pn.1) presents in section an oval shape, consisting of columnar cells radially arranged and containing a cavity of an irregular form. The histological structure of the Anlage is as compared with that in fig. 3, more or less loose1), and the Anlage itself is thereby also distended. The lateral plate (lm.) shows, however, no marked progress. The left side of this figure and the right of figs. 11 and 12 represent the section through the fifth myotome (mt.V) and the Anlage of the pronephros (a.pn.2) for that somite. The Anlage presents almost the same development as that just described. The left half of fig. 12 and the right half of fig. 13 shows the sixth somite (mt.VI) and the Anlage belonging to it (a.pn.3). It can be inferred from the arrangement of its component cells that the Anlage has been just constricted off from the myotome, as is shown by the fact that the cells at the point marked with x of the visceral and parietal layers are not yet rearranged to form a continuous laver,—a condition which is observed not infrequently in younger embryos. Fig. 14 shows on the right side a section through the hind wall of the sixth myotome; the Anlage beneath it (a.pn.3) is, therefore, the hind part of that represented on the right side of fig. 13: it is entirely cut off from the myotome (mt.17), and the two layers at this point have completely fused together, enclosing a comparatively wide cavity. The same condition is observed in the

<sup>1)</sup> When the pronephric Anlagen are cut off from the myotome, their structure is at first loosened, that is, their component cells become loosely set together. Later the cells multiply themselves, and are again compressed by mutual pressure; giving a compact structure to the Anlage—probably the same condition observed by VAN WYHE in Selachian embryos ('89, p. 476).

section through the anterior border of the somite. This phase of constriction is doubtless earlier than that shown in fig. 3. The right half of fig. 16 and the left of fig. 14 is from the section through the mid-plane of the next following somite, *i.e.* the seventh. The Anlage of the pronephros (a.pm.4) is not yet cut off from the myotome (mt.VII), but the process is beginning as shown by a slight constriction and an inclination to arch out, while the suddenly weakened lateral plate (lm.) forms the distal (ventral) continuation of it. This feature of the mesoblast reminds us of the youngest stage of the Anlage described above (compare with figs. 2 and 5). In the section passing through the anterior or the posterior border of the somite too, the same condition of the Anlage, as on the right side of fig. 14, is observed.

From the facts mentioned above, it is easily understood that the separation of an Anlage from a myotome begins with the constriction which takes place at the anterior and the posterior border of the somite, and that the middle portion is the last to be cut off, so that the cavity of the myotome (myocoelome) communicates with the peritoneal cavity, during some time, through a narrow passage at the middle point.

Myotomes when cut off from the Anlage of the pronephros assume a pentagonal form (see fig. 3) constructed of a dorsal, a lateral, a ventral, and two median sides, each of which is composed of a regular row of tall cylindrical cells. The dorsal and lateral rows of cells constitute the parietal layer of the myotome, while the three other sides represent the visceral layer (compare with the description on p. 314).

For about ten segments behind the seventh somite, the Anlage of the pronephros shows the same condition as that seen on the right side of fig. 16. Fig. 17 represents a section

through the twelfth somite; we can find no marked difference between the Anlage in the seventh somite and in this. The segments lying still farther backwards are not cut through exactly transversely in this same series of sections, owing to the cause stated above (pp. 312 and 315), so that we can not trace the differentiation of the mesoblast from the anterior to the posterior part in this one series. But I could demonstrate from several other series of sections that the Anlage of the pronephros is, in the present stage, found in no less than 15 somites.

Figs. 9-11 represent the contiguous sections through the intersomitic portion, on the right side, between the first and second Anlagen, i.e. between that of the fourth, and that of the fifth, somite. Fig. 9 is from the section next behind that shown in fig. 8; the portion (cd.) lying proximal to the lateral plate (lm.) presents no longer a weak appearance as in younger embryos (see the statement on p. 317 and figs. 4 and 7), but is occupied by a compact cellular structure (cd.) which suddenly passes over into the loosely composed lateral plate (lm.). Fig. 10 is from the section next posterior to fig. 9 and next anterior to the second Anlage represented in fig. 11 and shows almost the same condition as in fig. 9, with respect to the structure in the proximal portion of the lateral plate. In other words, in the intersomitic portion between the first and second Anlagen, a cellular cord has become established, which connects these two Anlagen. It is this cord which gives rise to the collecting duct or Sammelrohr of Rückert ('88), putting all the pronephric tubules in communication.

On the left side of figs. 9, 10, and 11, the contiguous sections through the intersomitic portion between the Anlagen second and third, are represented. In figs. 9 and 11, the condition of

the structure (cd.) at the proximal portion of the lateral plate is almost the same as that on the right side just described, although it is here somewhat weaker in development than there. The section represented in fig. 10 intervenes between the two mentioned above; in this section, the structure in question (cd.) is weakest in development, consisting of four or five cells only. In the left half of fig. 15 which represents the section through the intersomitic portion between the sixth and seventh somites, there is found no structure to be compared with the cord mentioned above, the proximal edge of the lateral plate (x) being of the same condition as that in figs. 4 and 7. In fact, the cord appears after the complete separation of the Anlage from the myotome, and when it is first established, the nearer the plane of a section to the Anlage either anterior or posterior, the thicker the cord. For instance, of the above three sections (the left side of Figs. 9-11), the middle (fig. 10) is the weakest. But this unequal development of the cord is soon made even by its growth as seen in the case of the cord between the Anlagen first and second (the right side of figs. 9 and 10).

The history of this cord as given above shows that it has doubtless the genetic relation with the Anlage of the pronephros. In early stages, no such structure is found in the intersomitic portion, but it becomes established one after the other with the development of the Anlagen. The cord is in section, thickest near the Anlage and weakest in the midway between two consecutive Anlagen, when it is first established. These facts give naturally an impression that it is growing out of the two consecutive Anlagen backwards and forwards and these two growing ends meet at some point in the midway between these two Anlagen, finally to fuse together. This point of meeting is, I think, indicated by the part where the duct has been described above as weakest. It is also the fact that repeated cell-multiplication takes place at the outer rim of each Anlage of the pronephros. One might suppose that the product of the cell-division would contribute only to the growth of the Anlage itself and has nothing to do with the cord; but this is not the case: the Anlage does not grow at the outer (lateral) end, as it might seem, but by cell-division within its own structure. I have never observed any case of cell-proliferation along the dorsal edge of the lateral plate in an intersomitic portion, although the cords appear, in later stages, to have some connection with that edge, when they are fully established (see the right side of figs. 9 and 10); this connection thus is not primary, but secondary. The epiblast has, from the first, no share in the formation of the cord, always showing a sharp contour against the mesoblast below.

There is thus no difficulty in accepting the rice that the connecting cord is formed of the intersomitic cell-outgrowths which are budded out of the anterior and posterior rims of each Anlage of the pronephros and are subsequently fused together. The cord is, therefore, originally brought about by the confluence of the free extremities of the Anlagen.

Further development of the Anlage of the pronephros may be intelligible by refering to fig. 18 which represents a section through a little older embryo of Stage II. The epiblast (ep.) consists of a single layer of cubical cells as before; the neural cord (n.) is still solid. On the left side of the figure, the hind border of the fourth myotome (mt.II) is cut, while on the right, the mid-plane of the fifth myotome is met with. A comparison with the corresponding parts in the younger stages (figs. 3 and 8) will

324 s. hatta :

plainly demonstrate a progressive change undergone by the pronephric Anlage. The Anlage on the right side (a.pn. 2) presents a feature much like that seen in fig. 3, notwithstanding some points of progress. The Anlage on the left side (a.pn. 1), however, shows a considerable progress: it has become much more compact by the active multiplication of its component cells. Owing to mutual pressure, the cells are compressed and their nuclei are regularly arranged, describing together an ellipsoidal figure. The inside of the ellipse encloses a comparatively large lumen, which is standing in connection with the body-cavity represented, at the present stage, only by the boundary line of the parietal and the visceral layer of the lateral plate (lm.).

In a little more advanced embryo, the cross-sections of which are represented in figs. 20-31, the neural cord (n.), the myotomes (mt.), and the Anlage of the pronephros show some progress as compared with those described in the preceding pages. The epiblast (ep.) consists, as in the embryo just described, of a single layer of cubical cells and is limited by a sharp line against the structures below. The component cells of the neural cord become arranged in two layers, leaving, in the anterior section of the cord, a vertical fissure-like lumen in the median line of the cord, which represents the beginning of the central canal (figs. 20-21, &c.). The posterior part of the cord is still solid, although the position of the central canal is marked by a vertical line produced by cell-boundaries (fig. 26) just as described in the foregoing pages. The myotomes are, in the anterior region, likewise enlarged, probably owing partly to multiplication of component cells and partly to

<sup>1)</sup> Owing to the same cause as the sections represented in Figs. 2 and 3, the vertical diameter of the neural cord in Figs. 20:23 is shown somewhat longer than it is in reality.

the loosening of the composition of the tissue<sup>1)</sup> and assume the shape of a scalene triangle (figs. 20-23); the median side of the triangle (mus.) represents the visceral, and the two other sides (cut.) the parietal, layer of the myotome. In the posterior region, they are yet of a compact structure of a pentagonal form, enclosing a eavity (figs. 25-31, mt.VII-X).

The anteriormost Anlage of the pronephros is found as before under the hind part of the fourth myotome, the section of which is represented in fig. 20 (a pn. 1). It shows a considerable development: the component cells, which are of high columnar character are no longer compressed, but the tissue is more or less loosened. Thus the Anlage itself is distended, and its upper (dorsal) angle becomes acute and grows in between the epiblast and the myotome. The internal cavity of the Anlage also becomes conspicuous. The Anlage of the pronephros under the next posterior myotome (the fifth) is not so advanced as in the last somite (the fourth). In fig. 22 is shown the section through the hind part of this somite and of the pronephric Anlage belonging to it (mt.V and a.pn.2), a section through the mid-plane being unfortunately wanting in this series of sections. The next posterior Anlage is found just under the sixth myotome and represented in fig. 24 (a.pu.3) together with the hind border of the myotome (mt.VI). The Anlage shows a compact structure which is probably due to a rapid multiplication of the constituent cells. The next following Anlage of the pronephros is found beneath the seventh myotome (fig. 26, a.pn.4). It shows no further development than the separation of it from the myotome and the fusion at the retrenched ends of the two layers of mesoblast: it is in the same stage of constriction as that in the

<sup>1)</sup> See the foot note in p. 319.

326 s. hatta :

right of fig. 13 which represents the Anlage third in a younger embryo.

The Anlagen above referred to are connected with each other by the solid connecting cords (figs. 21, 23, and 25, cd.), which are found between these Anlagen just as described in the younger stages. Of these connecting cords, that between the Anlagen second and third (fig. 23, cd.) is the thickest, while that between the Anlagen third and fourth (fig. 25, cd.) is the weakest in development, owing probably to its having been just established. The cord between the Anlagen first and second (fig. 21, cd.), however, is rather weaker as compared with that in the next posterior intersomitic plane (fig. 23, cd.). Such a case is occasionally met with: but this is doubtless not normal; in most cases examined, the cord is thickest in anterior segment and decreases in thickness gradually posteriorly as seen in the last example (see pp. 321-322 and figs. 9-11).

The Anlage of the pronephros belonging to the eighth somite and that of the ninth somite are not completely cut off from the myotome to which these respectively belong (figs. 28 and 30). They are, however, constricted already at the anterior and posterior borders of the segment: fig. 27 shows the posterior part of the Anlage fifth, and fig. 29 represents the anterior part of the Anlage sixth respectively. Such a case is observed in the younger stage described in the foregoing pages (pp. 319-320). Compare these two figures with the right half of Fig. 14 and the description on p. 319: here the phase of constriction is a little more advanced than there. The anterior part of the Anlage fifth and the posterior part of the Anlage sixth, the figures of which are omitted, have features much like those seen in figs. 27 and 29. In these two segments, the central portion of the Anlage is

just in the process of being constricted off from the myotome, and we can not decide by this case alone which segment (whether the anterior or the posterior) is the further developed; a comparative study of other examples shows that the separation in the posterior segment follows that in the anterior. The state of the mesoblast in the next posterior segment, i.e., the tenth segment (fig. 31), is quite different from that just described; it is in a more primitive condition of development. The Anlage of the pronephros (a.sd.) presents only an indication of constriction,—a feature which we have observed repeatedly in embryos of younger stages (compare with figs. 2, 5, 6, 14, and 16). From this segment backwards, a few segments show almost the same condition. Still further posteriorly, the structure of the mesoblast can not be readily observed, since the planes of sections incline by degrees in the cranio-caudal direction, owing, as above stated, to the bending of the longitudinal axis of the embryo.

In all the segments mentioned above, the lateral plate (lm.) consists of a loose tissue of cells of variable shape, and the Anlage passes over suddenly into the lateral plate just as in the embryos described in the foregoing pages.

In this stage, therefore, the Anlage of the pronephros is completely separated from the myotome in 4 somites, i.e., from the fourth to the seventh inclusive; and these are connected with one another by the intersomitic solid cord. In the following 4 or 5 somites, the constriction is just going on while in a few of still more posterior somites it is indicated merely by a slight depression in the parietal layer of the mesoblast.

#### Period 2.

In the embryos which belong to Stage III, we observe a decided advance in several respects. Figs. 32-50 represent a series

of cross-sections through one of these embryos which has about 25 somites. The neural cord (n.) is reduced in size and in the anterior part has a conspicuous canal. The myotomes which showed before a pentagonal outline, in the anterior part of the body have now assumed an elongated lozenge-shape<sup>1)</sup> and is composed of an outer, and an inner, layer of long cells which have begun to diffentiate themselves. The inner layer (mus.) represents the median and ventral rows of the pentagonal myotome mentioned on p. 320 and, therefore, corresponds to the visceral layer of the myotome; the outer layer (cut.) is the product of the dorsal and lateral layers and constitutes the parietal layer. The pronephric Anlage is composed of high columnar cells which are plainly distinguishable from the much shorter elements of the lateral plate (lm.). component cells of the Anlage of the pronephros which we generally found to be compressed in the foregoing stage (pp. 324 and 325), are now more or less loose, and the internal cavity of the Anlage is somewhat widened, being distended by the loosening of the cells.<sup>2)</sup>

The peritoneal cavity is, at the present stage, still represented merely by the boundary-line of the parietal and visceral layers of the lateral plate.

In the present stage, the foremost Anlage of the pronephros is, as before, found under the hind part of the fourth myotome (figs. 32-35, a.pn.1). The Anlage shows, in section, a circular outline and is composed of high columnar cells arranged in a radial manner. The internal cavity of it is confined no longer to one section, but it is observed in three or more sections; it is most spacious in the hind part of the fourth somite (fig. 33) or in that part where the cavity is visible from an early period.

<sup>1)</sup> A few myotomes in the anterior somites tend to assume this shape already in the last stage (see figs. 20, 21, and 22).

<sup>2)</sup> See the foot-note on p. 319.

From this part backwards it gradually decreases in width until no space is perceptible. Anteriorly the cavity is also somewhat narrowed, but not as much as in its posterior continuation, and ends blindly rather suddenly at its anterior end (fig. 32). The anterior portion of the Anlage forms a blunt conical tube (Fig. 32, a.pm.1) projecting anteriorly and lying between the dorsal edge of the lateral plate (lm.) and the lower surface of the fourth myotome (mt.IV). The existence of this conical tube<sup>10</sup> gives us a strong impression that originally there must have been present an Anlage of the pronephros in the anterior segment which was connected by a connecting cord with the Anlage belonging to the fourth somite, but had disappeared during the phylogeny and that this conical tube is the remnant of this connecting cord.<sup>20</sup>

The next posterior Anlage, which is found under the fifth myotome (figs. 37-39, a.pm.2) and shows an outline much resembling that represented in figs. 32-35, has an internal cavity of irregularly triangular form, extending through three sections, of which the foremost section contains the most spacious cavity, while in the others the lumen grows smaller and smaller. The pronephric Anlage in the next following somite (figs. 41-43, a.pn.3) has an outline much like that shown on the left side of Figs. 18 and 24, being in the same phase of development, that is, it is of the form of an isosceles triangle whose two basal angles touch the myotomes. This Anlage is found under the sixth

<sup>1)</sup> The internal cavity of this conical tube is not entirely closed, but there is clearly seen a small canal (x) directed towards the median side and opening below the myotome. I can not decide, at present, whether this canal is normal or abnormal; for 1 can not make out the corresponding structure on the opposite side and have no other embryo of exactly the same stage, in which the structure in question would probably be found, it it be of some definite meaning; 1 also can not detect any trace of such a canal in embryos of advanced or younger stages.

<sup>2)</sup> See the description under Period. 4.

330 s. hatta :

myotome and contains the internal cavity extending likewise for three sections, of which however the hindmost contains the widest eavity, while it is diminished in width anteriorly: in other words, the width of the cavity enlarges in inverse direction as compared with that in the preceding two somites. The Anlage fourth under the seventh myotome (figs. 45 and 46, a.pn. 4) has an oval outline like that shown in fig. 26 and encloses an internal cavity, which covers two sections and is anteriorly wide and posteriorly narrow. The two following Anlagen which are detected under the eighth and ninth myotomes respectively (fig. 48, a.pn. 5 and fig. 50, a.pn. 6) show almost the same condition of development as in the somite just described; the internal cavity which they contain is likewise extended into two sections; the width of the cavity is about the same in these two sections, being of a fissure-like form.

The solid cord which is observed in the embryos of the last stage connecting the Anlagen with one another, is also found here. The cord in the intersonitic plane between the Anlagen first and second (fig. 36, cd.), that between the Anlagen second and third (fig. 40, cd.), and that between the Anlagen third and fourth (fig. 44, cd.) are all comparatively short, so that they are in each stretch confined to only one section, while that in the two posterior intersonitic planes, i.c. between the Anlagen fourth and fifth, and between the Anlagen fifth and sixth, the cord is extended in each case into four sections. In this latter part, the cord is in a primitive condition; the component cells are actively multiplying. Hence these four sections all show similar features. I have endeavoured to show in fig. 47 one of these sections which is taken from one of the four sections between

the Anlagen fourth and fifth, and in fig. 49, one between the Anlagen fifth and sixth.

This inequality in the length of the intersomitic solid cord is, I believe, due to differences in the degree to which the canalization within the Anlage has extended into the connecting cord. In the anterior section of the pronephros, this process has already proceeded to some extent into the interior of this cord, while in the posterior, the cavity is still confined entirely within the Anlage itself. The whole system of the pronephros at the present condition may be compared to a bamboo-cane with nodes and internodes; in the anterior section of the system, the nodal septum has become very thin, while it has a considerable thickness in the the posterior. As will be shown further on, all these septa entirely disappear later when the collecting duct is fully established.

From the fact mentioned above, it will be easily seen that the process of canalization in the pronephric system of *Petromyzon* begins in the internal cavity of the pronephric Anlage in each segment and is extended into the intersomitic connecting cord. The direction in which this process proceeds seems, generally speaking, to be from the anterior section to the posterior; for in most cases, not only the internal cavity in each Anlage is spacious anteriorly and narrowed posteriorly, but the cavity in anterior somites is extended more, or canalization goes on further, than in the posterior section of the system; although the progress in the opposite direction is occasionally met with.

From the tenth somite backwards, five or six segments show the same condition of the mesoblast as in the eighth and ninth somites, after which the series can not be studied, owing to the inclination of the planes of sections, referred to above. In all the segments above referred to, the lateral plate of the mesoblast shows the same condition as in the foregoing stages, but has become more distinct from the Anlage of the pronephros.

In the present stage of development, then, the Anlage of the pronephros is ent off from the myotome in more than 10 segments, and the canalization has advanced in the anterior section of the system, to a state just ready to put the Anlagen in the succeeding somites in communication with one another, although the intersomitic connecting duct in the posterior part remains still solid.

Figs. 51-58 were drawn from a series of sections through one of the older embryos in this stage. The internal structures are developed much more than in the embryo just described. The cells forming the visceral layer of the myotome have been differentiated into the muscle-plates, while the parietal layer is composed of cubical cells. The Anlagen of the pronephros have acquired, in most cases, a tubular structure and have grown dorsally, being folded out from the body-cavity: I will accordingly call them the pronephric tubules.

On the right side of fig. 51, the foremost tubule ( $\rho t.1$ ) is visible, which no longer contains the internal cavity but is converted into a solid mass of cells occupying the space beneath the fourth myotome. This consolidation is not due to retrogressive changes, but is effected by very active cell-multiplication which takes place within the tissue. The cross-section of the collecting duct seen on the left side of fig. 52 (cd.) which represents the third section behind the last, is likewise solid. The tubule on the right side of this figure ( $\rho t.2$ ) and that on the left side of the third section posterior to it (fig. 53,  $\rho t.2$ ) are respectively the second tubule of the right and left side found

under the fifth myotome; both are of a triangular form and contain a very spacious internal cavity of the same shape. On the right side of fig. 53, the sixth myotome and the third tubule are shown. The section next posterior to fig. 53 (fig. 54) shows the cross-section of the collecting duct (cd.) on the right side and a slice of the hind wall of the second tubule on the left (pt.2). The cells composing the duct are closely set together, although arranged more or less radially, acquiring a tubular form. has been repeatedly mentioned above, the epiblast is, as in the foregoing stage, marked off from the mesoblast as well as from the Anlage; but at the present stage, the second tubule (figs. 54, pt.2) pushes against the epiblast, probably in consequence of an enormous multiplication of its component cells, so as to cause the latter to be a little elevated externally. It must be remarked here that the Anlagen, especially the first and the second, when they first assume the tubular form, are brought into an intimate relation with the epiblast, striking against it. In some of my sections, a mitotic figure is seen at that point of the epiblast<sup>10</sup> (fig. 54, x). This might lead some to assume a genetic connection between the epiblast and the pronephros in Petronyzon; but there is, I believe, in reality no such relation. If the epiblast gives some cells destined to build the pronephros or a part of it, cell-proliferation or some other mode of cell-production would necessarily be observed in the epiblast in the preceding stages or at least, in the stage here spoken of. In the foregoing stages, the epiblast had, as has been repeatedly mentioned above, a sharp limit against the structures inside it. At the present stages also, it is marked off by the boundary-line of cells from the tissue of the tubule,

<sup>1)</sup> In the series of sections, from which fig. 54 is drawn, I observe mitotic figures at that point in several sections.

showing no structural alteration. Mitotic figures are met with not infrequently in that part of the epiblast (fig. 54, x); their axis lies, however, in all the cases examined parallel to the plane of the epiblast, giving us an impression of the resulting cells contributing to the formation of no other part than the epiblast itself; on the contrary, within the structure of the tubule the cells are rapidly multiplying (figs. 51, pt.1 and fig. 54, pt.2), showing that the growth of the tubule is actively going on. In fact, the connection, or rather the intimate contact, of the pronephric tubule with the epiblast is a temporary condition; the separation follows immediately afterwards, and the tubule returns soon into a state similar to that seen in fig. 52 (pt.2).

According to RÜCKERT ('88), a similar case is observed in Selachian embryos: the tubules become connected secondarily with the epiblast—what caused him to believe that the latter might give some constituent elements to the tubules.

The third section behind that represented in fig. 54 (fig. 55) shows, on the right side, the fourth (pt.4) and, on the left, the third tubule (pt.3) respectively. The latter is not so far developed as its counterpart on the opposite side (fig. 53, pt.3), while the former presents a great progress: it consists of a definite epithelium and contains a distinct cavity of triangular shape, although the corresponding tubule on the opposite side (fig. 56, pt.4) which is found in the third section behind the last, is much less advanced in development. The fifth tubule, the tubule on the right side of fig. 56 (pt.5), is somewhat more developed than that which belongs to the anterior somite (the fourth tubule on the opposite side); but it has a feature much resembling the fourth tubule on the same side (fig. 55, pt.4) and the second on the

opposite side (fig. 53, pt.2). In short, in this series of sections, the tubules on the right side, are all more advanced than those on the opposite side. The sixth is very primitive in development; fig. 57 represents the section, on the left side, through the anterior part of the ninth somite and, on the right, the posterior part of it. The left tubule is sliced at its anterior wall, but the right tubule is cut through in its mid-plane. It is composed of two layers of columnar cells, but no cavity has yet appeared in the interior.

From the tenth somite backwards, the Anlagen are cut off from both the myotomes and the lateral plate, and constitute the segmental duct or the posterior continuation of the collecting duct, which is distinctly traceable for 7-8 somites. Not infrequently, however, a somite is met with, in which the segmental duct is not yet cut off from the lateral plate at the time when the separation is finished in a majority of somites, as seen in fig. 58 which represents a section through the twelfth somite. The left half of the figure shows the duct entirely cut off from the lateral plate, while the right exhibits the state not yet separated. The same structure is made out in two contiguous sections, so that one might mistake it for a pronephric tubule. This point will be described further on.

The relation of the pronephric tubule and the peritoneal cavity is not so simple as in the last specimen; besides the pronephic tubule, there is seen another structure which projects out of the inner angle of the peritoneal cavity (figs. 52, 53, 55, and 56, c.p.). This projection is originally a fold of the peritoneal wall and gives rise, as subsequent history shows, to the radix of the mesentery, whence the gonads and the mesonephric tubules are derived. It will here be called briefly the "coelomic projection."

At that point of the visceral layer of the mesoblast, where the Anlage of the pronephric tubule passes over to the lateral plate, it is always many cells deep (figs. 55 and 56, c.p.), and the projection in question is brought about by repeated division of these cells. The projection formed is consequently seen in each somite and thus shows a segmental arrangement. Its component cells are soon re-arranged into an epithelium, and the pouches thus formed push their way between the myotome and the hypoblast.

The coelomic projection appears, at first sight, to be homologous with the coelomic pocket described by Price ('97) in Bdellostoma. The coelomic pocket is, however, according to Price, the product of both the parietal and visceral layers of the lateral plate and is afterwards converted into the cavity between the glomerulus and Bowman's capsule of the Malpighian corpuscle; the floor of the pocket forms Bowman's capsule, and its roof together with a part of the pronephric tubule is transformed into the cover of the glomerulus ('97, p. 213). The coelomic projection in Petromyzon is, on the contrary, formed out of the visceral layer of the distal half of the somite and gives rise, as just stated, to the radix of the mesentery, from which partly the mesonephric tubule and partly the gonads are formed.<sup>10</sup>

Figs. 59-63 are from a series of sections through an older embryo of the same stage. In this series of sections, a further development of the coelomic projections is clearly seen; the first figure (fig. 59) shows the section through the second tubule, fig. 60 through the third, and so forth. In the first 3 figures and on the right of fig. 62, the coelomic projection (c,p) presents an

<sup>1)</sup> I will not here further discuss this structure, as I intend to do so in a future paper in which the development of the mesonephros in Petromyzon will be delt with.

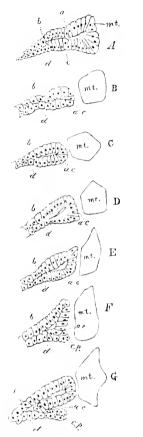
epithelial structure, forming the continuation of the peritoneum and folding out from the peritoneal cavity. Beneath the first tubule, there is found no rudiment of the projection: under the second (fig. 59) it is very weak, while beneath the third (fig. 60), fourth (fig. 61), and fifth (fig. 62), tubule, respectively it is most vigorously developed. But on the left side of figs. 61 and 62 it is again in a primitive condition, just as in the last series of sections (figs. 52, 53, 55, and 56).

The coelomic projections are not confined to the anterior region where the pronephric tubules are found, but it is found likewise in the posterior part where only the segmental duct develops. Fig. 63 shows the section through the thirteenth somite; on this section, the duct is cut off from the myotome and a well developed coelomic projection (c.p.) is observed; I will return once more to this subject further on.

Leaving the coelomic projection in this stage of development, I will return to the origin of the Anlage of the pronephros and give somewhat more exact details on the subject. Since the piece of the mesoblast called above the Anlage of the pronephros forms for a time the proximal portion of the lateral plate, one might presume that its whole mass will be transformed into the pronephric tubule and will not partake in the formation of the peritoneal membrane. I was at first of this opinion, but a careful observation of sections through the embryos in each stage showed my error.

To illustrate this point satisfactorily, I have given, in the annexed wood-cut (Wood-cut 1), a series of semi-diagramatic figures, which show the successive phases of the changes going on in the structure. A shows the first indication of the Anlage of the pronephros before the separation of it from the myotome;

a-b indicates the extent of the Anlage; c-d shows that of the coelomic projection. When the myotome is cut off, the point



Wood-cut 1.—Semidiagramatic figures to illustrate the successive phases of the evolution of the nephrotome.

- .1. from the right side of fig. 16.
- $B_*$  from fig. 3.
- C. from the right side fig. 18.
- D. from the left side of the same.
- E. from the left side of fig. 53.
- F. from the right side of fig. 55.
- G. from the left side of fig. 61.

of the parietal laver indicated by a becomes fused with the point a of the visceral layer (B, ac). This piece of the mesoblast assumes an ellipsoidal shape (C). The component cells of this ellipsoid are multiplied by active cell-divisions, and the piece almost loses its lumen and gets a compact consistence (D). Meanwhile the cells in the space d-c remain inactive. Consequently the piece acquires a triangular form (E), whose upper sharp angle, together with the two sides enclosing this angle, gives rise to the pronephric tubule. The lower (median) obtuse angle now begins to grow by cellmulplication (F) and produces a small knob (F, c.p.), which grows further and pushes in between the myotome and the hypoblast (the upper wall of the This cellular enteric canal). projection is that which has been

called the coelomic projection. It is reduced into a thin plate of epithelial cells (G, c.p.) and assumes then the form of

a true fold of the visceral layer of the lateral plate. At the same time, the upper angle or the pronephric angle develops further and assumes a tubular form composed of a single layer of columnar cells (G).

The peritoneal cavity begins, therefore, at the point, from which the coelomic projection starts, and the part of the layer dorsal to this point is all appropriated to the formation of the pronephric tubule. The nephrostome will be found, therefore, by the point where the tubule passes over to the projection.

I will add a few words on the differentiation of the myotome, so far as concerns the topographical relation of it to the Anlage of the pronephros. The invotome consists, at the present stage (Stage 111), of the inner and outer lavers which constitute respectively the Muskelblatt and the Cutisblatt of German authors (fig. 59 and 60, mus. and cut.). The cells composing the Muskelblatt (mus.) are, simply differentiated into a transverse row of the muscle-plates. The outer laver (cut.) undergoes, however, subsequently a series of interesting changes: it folds in, just as the Sklerablatt or sclerotome described by Hatchek in Amphiorus ('88) between the Muskelblatt and the chorda and the neural tube. 11 As is well known, Rable ('88) has homologised Hatchek's Sklerablatt with his Sclerotomdivertikel of Selachian embryos, which is the evagination of the ventral part of the visceral layer of the mesoblastic somite. This part of the somite (the selerotome) corresponds, I believe, exactly to the ventral row of the pentagonal myotome in my embryo (see pp. 314 and 320), which comes afterwards to form the ventral part of the cutis-laver (see figs. 21, 22, 23, 36, 37, 43, 49, 59, 60, &c.). When the invotome is not vet separated from the rest of the mesoblast (fig. 2), this part of the

I) This subject will be treated of in an independent article.

340 s. hatta:

selerotomic layer forms, as has been seen above, a direct continuation of the visceral layer giving rise to the coelomic projection. (The successive changes of the myotome are seen in figs. 1, 2, 3, 18, 21, 22, 43, 49, 60, &c.)

From the above account, it can be inferred that the ventral half of the mesoblastic somite in *Petromyzon*, which gives rise to the pronephic Anlage and the coelomic projection, is doubtless homologous with the "intermediate cell-mass" of Balfour described by him in Selachia and, therefore exactly coincides with the "Nephrotom" of RÜCKERT. So far as concerns its future destination, however, the results arrived at by me slightly deviate from their views.

A series of sections through the oldest embryo of this stage is represented in figs. 64-76. The epiblast has undergone no histological change, but remains, as before, one cell deep. Many structures, however, exhibit a remarkable progress. The muscle-layer (mus.) of the myotome is, for instance, further differentiated, now consisting of a transverse row of long muscle-cells, although the cutis-layer (cut.) is still composed of short cubical cells. In the anterior region, the true coelome (pp.c), becomes conspicuous enclosed by the parietal (m.p.) and the visceral (m.r.) layers of the lateral plate, both of which consist of a single row of cubical cells. The ventral edges of the lateral plates on both sides do not, however, yet meet in the ventral median line. The walls of the enteric canal too are, in the anterior region, reduced into a single cell layer.

A great alteration is met with in the pronephric tubules. They have assumed a cylindrical form composed of tall columnar epithelium and have grown dorsally, pushing in between the myotome and the epiblast, causing the latter to be elevated a

<sup>1)</sup> In spite of the discussion by RÜCKERT ('89, pp. 19-20) on the inexactness of the expression "intermediate cell-mass," I homologise, with many authors, these two terms with each other.

little. The internal lumen of the tubules are put not only in wide communication with the peritoneal cavity, but also in direct continuation with one another through the collecting duct, which consists of a regular columnar epithelium-cells arranged radially and now encloses a conspicuous lumen.

In the foremost of these twelve sections (fig. 64), we notice that a structure (pt.1) consisting of a few cells projects at the outer corner of the proximal edge of the lateral plates and lies in contact with the outer wall of the myotome on either side. This structure is found under the anterior border of the fifth myotome and I infer that it is a remnant of the first pair of the pronephric tubules which begins to decline in the present stage. The reason why it is found not under the fourth myotome as in all the stages hitherto described but beneath the anterior border of the fifth myotome, consists probably in its shifting backwards; for we find, in this series of sections, another pair of the tubules under this same fifth myotome. A comparison of this figure with fig. 65 representing the next posterior section will make the matter clear. On the left side of fig. 65, the same remnant structure (pt.1) together with the collecting duct (cd.), which connects the first and the second tubules can be observed, while the cross-section of the collecting duct in the corresponding intersomitic plane is seen on the opposite side (cd.). The next following section is shown in fig. 66; the tubules (pt.2) on both sides communicate freely with the peritoneal cavity; these are found beneath the hind part of the fifth myotome and are the second pair of the tubules; the openings (nst.2) to the peritoneal cavity are, therefore, the second nephrostome of the pronephros. The tubule on the left side is weaker than that on the right, since a larger part of the left tubule is visible on the section next posterior which is represented in fig.

67 (pt.2). The shape, which the tubules of the second pair (fig. 66, pt.2) assume at about this stage, is a characteristic triangle, whose two angles, the one directed dorsally and the other directed medially, are acute and whose outer (lateral) angle is obtuse (see the left side of fig. 59 and the right side of fig. 66, pt.2); so much so that we can easily determine by this feature the fact of their being the second pair. This peculiar shape of the second tubule is retained for a considerable time as will be seen further on. On the right side of fig. 67, the collecting duct (cd.) is cut through transversely; on the left, the same duct (cd.) and the hind part of the second tubule (pt. 2) are seen.

At the point where the nephrostome opens to the peritoneal cavity, the visceral peritoneum at the median corner of the latter projects out between the myotome and the hypoblast (figs. 66 and 67, c.p.); beneath the collecting duct, however, no such structure is detected (see the right side of figs. 65 and 67). Such a ponch is repeated in each nephrotome (see figs. 66-75, c.p.) and is what has been called above the coelomic projection.

The next following sections shown in figs. 68 and 69 show the third pair of the tubules (pt.3) to be of the same structure. In these two sections the tubules are cut through lengthwise, and the nephrostomes (nst.3) on the two sides come into view in symmetrical manner. The tubules are so simple as to need no further explanation. Fig. 70 represents a section through the intersomitic plane between the sixth and the seventh somites, and next posterior to fig. 69. It shows on either side the cross-section of only the collecting duct (cd.), consisting of radially arranged cells. Fig. 71 is from a section through the seventh somite and is the third

<sup>1)</sup> In this series of sections, we often see the coelomic projection on sections passing through the intersomitic plane; but this is the piece of it belonging to either the anterior or the posterior somite.

behind the section shown in fig. 70; the tubules of the fourth pair (pt.4) show themselves symmetrically on both sides; they are somewhat less developed as compared with those of the last pair. Fig. 72 is the section next behind fig. 71; it shows on both sides the collecting duct (cd.) together with the coelomic projection (c.p.)which is a part of that of the anterior segment. Fig. 73 is the third section posterior to that just described; the blastoderm becomes more flattened than in the foregoing sections; it shows on both sides the tubules of the fifth pair (pt.5); the condition of the tubules and nephrostomes (nst.5) is much like that in fig. 71. Fig. 74 is from the fourth section posterior to fig. 73; the right tubule (pt.6) of the sixth pair and its nephrostome (nst.6) are visible on the right side, while the collecting duct (cd.) is cut through on the The sixth tubule and nephrostome on the opposite side are observed in the next anterior section which is not figured. The segments back of the ninth somite have no trace of the tubule, but the cross-sections of the posterior continuation of the collecting duet, the segmental duet, are repeated in each section. Fig. 75 represents a section through the sixteenth somite; the crosssection of the segmental duct (sd.) on either side is seen; it always occupies the space where, in the anterior region, the tubules or the collecting duct is found.

This condition, however, is not continued to the dorsal lip of the blastopore. As I have stated in my previous paper ('91), many processes of development are much delayed in the hind region, so that we are here reminded of what were seen in the anterior region of the younger stages. Fig. 76 represents the fifteenth section from the dorsal lip of the blastopore and passes through about the twenty-third somite. In this comparatively late stage, in which many mesoblastic organs have developed in the anterior

region, the neural cord (n.) is still solid; the mesoblast (ms.) is many-cell-layered and its metameric segmentation is still going on. On the right side of the figure, the section passes through the mid-plane of the myotome showing no sign of its separation from the rest of the mesoblast, while on the left, which shows the intersomitic portion, the process of separation (mt. and lm.) is going on. On both sides, however, there is no structure that can be recognised as the Anlage of the segmental duct.

The six pairs of pronephric tubules observed in this stage are the maximum number for Petromyzon; this stage ought, therefore, to be regarded as the highest point of development with reference to the pronephros. Even in the present stage, the foremost tubules show a tendency to degenerate.

### Period 3.

The embryos of Stage IV, which have about 35 mesoblastic somites, present a remarkable progress. The head-fold is much prolonged: in older embryos of this stage, it begins to twist ('97, fig. 1, D). Figs. 77-91 represent sections through one of these embryos. In some myotomes, the selerotomic fold goes deeper between the muscle-layer and the chorda. The parietal layer of the lateral plate (m.p.) is much lessened in thickness, so that it is reduced, in the dorsal region (the posterior two thirds of the pronephric extent), into a thin epithelial lining of the body wall (see figs. 79-85). The coelomic projection is likewise reduced into a thin plate (figs. 82-86, c.p.) except in the anterior two segments of the pronephic region, in which it still keeps the characters of the younger stages (figs. 77-81, c.p.), only folding in deeper than in the foregoing stages. The visceral layer (m.v.) of the

peritoneum still consists of a cubical or rather cylindrical epithelium. The pronephric tubules are, in general, much prolonged
and begin to coil in the dorso-lateral direction, so as to cause an
elevation in the epiblast. The walls of the tubules consist of a
regular row of cylindrical cells, which passes over suddenly into
the thin peritoneum (figs. 77-86), except in the region of the second
pair of the tubules, where the parietal layer (m.p.) of the lateral
plate still retains the character of the younger stages, being
composed of cylindrical epithelium like the tubules themselves
(figs. 77-79). At some regions, even a few mesenchyma-cells (mch.)
appear,—for instance, beneath the chorda (see figs. 80, 82, and 84),
in the median ventral space (see figs. 81 and 82), and also inside
the lateral epiblast (see figs. 77 and 80).

Fig. 77 shows a section through the fifth somite and therefore corresponds to fig. 66, which represents the section through the same plane of an embryo at a younger stage. The longitudinal section of the second tubule (pt.2), together with the corresponding nephrostome (nst.2), is seen on the left side of the figure, greatly resembling the tubules of the same pair in the younger stage (compare with fig. 66). On the right side, the nephrostome (nst.2) alone is observed; the tubule proper is to be seen in the two following sections which are represented in figs. 78 and 79.

Beneath the myotome anterior to the one just described, there is found neither a tubule nor any structure that may be regarded as the remnant of it. In the space between the epiblast, the myotome and the lateral plate, however, a few scattered cells (fig. 77, mch.) are found. I at first supposed that these might be disconnected component cells of the first pair of tubules; but, as free cells of quite the same character are found

in other places, for instance, in the space between the lateral plate and the epiblast (figs. 79-81, mch.), I have been compelled to conclude that they have no genetic relation with the proncphros, but are mesenchymatous cells which are destined to form the blood-vessels and corpuscles.

As the embryo was somewhat twisted, the sections did not pass through the lateral walls of the body in an exactly transverse plane, but unavoidably obliquely, on either side, as the continuous serial sections represented in figs. 79-86 show.

While on the left side of fig. 78 the posterior portion of the second tubule (pt.2) is seen, the second nephrostome (nst.2) is observed on the right together with a cross-section of a tubular structure (cd.). This latter might be taken as a slice of the anterior border of the second tubule, but is, in my opinion, the remnant of the collecting duct which once connected the second tubule with the first and forms, at present, a tubercle in front of the second tubule, the first tubule having disappeared; for the second tubule on that side is observable in the next following section represented in fig. 79 (pt.2) showing its characteristic features stated above (p. 342).

On the left side of fig. 79 and on the right side of fig. 81, we see the anterior half of the third tubule (pt.3) and nephrostome (nst.3) of each side, their respective posterior half being found on the left side of fig. 80 and on the right side of fig. 82 (pt.3 and nst.3); the tubules are bent laterally and dorsally, probably caused by the prolongation of their tubular portion; for their nephrostomal part and dorsal blind end retain their original position. This is the first step in the convolution of the pronephric tubule.

As seen on the left side of figs. 79 and 80, the tubule (pt.3)

of the third pair shows a new character: the dorsal blind end and the nephrostomal portion of the tubule are more or less expanded, while these two portions are united by a slender middle trunk. When compared with the tubule of the same pair on the opposite side represented in figs. 81 and 82 (pt.3), this character of the tubule will be understood more clearly: the dorsal expansion is seen in fig. 81, while the nephrostomal widening is observed in fig. 82. The tubules of the following two pairs show the same feature. On the right side of fig. 80, only the collecting duct between the second and third tubules is found. The left tubule of the fourth pair is shown on the left side of figs. 81 and 82 (pt.4); fig. 83 shows a cross-section through the collecting duct (cd.) between the third and fourth tubules and a slice of the anterior wall of the right tubule (pt.4) of the fourth pair which is prolonged and bent like the tubules of the last pair. The fifth pair of tubules is seen on the left half of fig. 84 on one side (pt.5) and on the right of fig. 85 on the other (pt.5). It is not developed as much as the more anterior pairs, but shows considerable progress as compared with the tubules in fig. 73 which represents the younger stage of the same pair.

These four pairs of the tubules (from the second to the fifth) contain a spacious lumen and stand in wide communication with the peritoneal cavity, which becomes, at the present stage, conspicuous from this region forwards.

In fig. 86 which shows the fifth section behind the section shown in fig. 85, the space on the left side which is occupied, in the more anterior region, by the tubule or the collecting duct, is replaced by the cross-section of a duct (sd.) with an oval outline and an ovoid lumen. This is the segmental duct under the tenth myotome (mt.X). On the right side of the figure, however,

besides the cross-section of a duct (cd.), there is seen a pronephric tubule (pt.6), the long axis of which is directed vertically to the inner surface of the epiblast. It is found just beneath the ninth myotome where the sixth tubule should be found. though the parts of it are also to be seen in two consecutive sections (the one represented in fig. 86 and another preceding it), the communication of its lumen with the collecting duct is not to be found anywhere. In some embryos, the tubule loses the connection with both the body-cavity and the duct. The structure (pl.6) in question is, I believe, nothing else than the remnant of the sixth tubule which is in a stage of degeneration, and the duct (cd.) is doubtless the collecting duct between the tubules of the fifth and sixth pairs. Compare the segmental duct (sd.) on the left side with the collecting duct (cd.) on the right side just described; the latter has a wide circular lumen, whilst in the former it is slender and compressed. This difference of character between these two ducts is noticeable for some time in the younger stages.

To sum up the results obtained in this stage the tubules of the third to the fifth pairs are rigorously developed, while the second is very weak, the sixth retrograding, and the first has entirely disappeared.

In the present stage, a peculiar structure is observed inside the walls of the body-cavity (figs. 77-85, pp.1-3). At some points of the peritoneum, a thin plate which consists, in cross-section, of one, two, or three cells, projects from the peritoneal wall into the body-cavity; it will be called here shortly the "peritoneal partition." A peritoneal outgrowth is found at the level where the coelomic projection passes over to the visceral layer of the lateral plate; at the same level, another peritoneal outgrowth starts up out of the parietal layer. These two outgrowths meet at midway

and cut off a long chamber from the body-cavity along the openings of the nephrostomes (figs. 77-85, pp.1). This longitudinal chamber communicates anteriorly as well as posteriorly with the body-cavity below, which is represented, in those parts, by the boundary of the parietal and visceral layers of the lateral plate. This is the uppermost partition. The second partition is weaker in development and is detected a little more ventrally, projecting likewise from the parietal and the visceral layers of the lateral plate. It is most obvious in the region beneath the third and fourth nephrostomes (81-84, pp.2). We find the third partition still more ventrally, which is weakest in development; its extent is almost the same as the second (figs. 81-84, pp.3).

These partitions disappear after a short existence; in a little older embryo, none of them is detected, as will further be seen. This is probably the same structure as the "peritoneale Scheide-wände" or "Peritonealbrücke" described by Goette in Petromyzon fluriatilis ('90). As to the meaning of the structure I have nothing to say."

It is important here to illustrate the topographical position of the pronephros and the relation of it to other parts; for these become definite for the first time in the present stage. For this purpose, a series of sagittal sections is instructive (figs. 112-114).<sup>2)</sup> A few anterior myotomes (mt.II-V) are seen in fig. 112, which represents the section nearest the median line. In the posterior part of these myotomes, four cell-layers are distinguishable; the outmost layer (ep.) is the epiblast; the cell cord (cd.) inside the epiblast is the longitudinal section of the collecting duct, and

<sup>1)</sup> See the historical review under Petronuzon.

<sup>2)</sup> The embryo, from which these figures are drawn, is a little younger than that just spoken of.

350 s. hatta :

its caudal continuation (sd.) is the segmental duct; while the inner two layers (m.r. and m.p.) present respectively the parietal and visceral layers of the lateral plate. Below these structures, the roof of the enteric canal covers the fore-gut (fg.) and the commencement of the mid-gut which forms the passage of the enteric cavity from the anterior slender portion to the posterior wide cavity. In fig. 113 which represents the section next outside the last, the lateral walls of the five myotomes from the first to the fifth (mt.I-I') are noticed; the cell-mass (au.) seen next anteriorly to the first myotome (mt.1) is a slice of the wall of the left auditory pit. The cross-sections of the pronephric tubules from the second to the fourth (pt.2-4) follow immediately behind the fifth myotome; an oblique section of the fifth tubule (pt.5) and the nephrostomal part of the sixth tubule (pt.6) are also obvious behind the fourth tubule. The nephrostomes of the second (nst.2) and fifth (nst.5) tubules are seen only in part, while a larger part of the third, fifth and sixth nephrostomes is seen in the next figure (fig. 114, nst.3, 5, and 6) which represents a section still further outside. The sixth tubule, which is of weak development and has a wide nephrostome, is visible in these two sections (figs. 113 and 114, pt.6 and nst.6). Except the first tubule which has already disappeared without leaving any trace, the five tubules are all thus seen in the dorso-lateral aspect of the hind section of the fore-gut and the commencement of the mid-gut.

The pronephros is thus situated in the neck which connects the head-protuberance with the globular abdominal portion. Below the pronephros, the narrow passage of the fore-gut passes through to unite the fore-gut with the wide space of the mid-gut, where afterwards the liver (l.) is found. Underneath the passage of the fore-gut, a group of mesenchymatous cells (mch.), which constitutes the earliest fundament of the heart, is detected. The present position of the pronephros,—dorsal to the heart, anterior and dorsal to the liver, and along either side of the chorda,—is retained by it for a comparatively long period (see fig. 97); in later stages, the liver is somewhat shifted backwards, so that the pronephros now comes entirely in front of it (see fig. 115).

In an older embryo of this stage (figs. 92-96) the median folds of the coelomic projection, the component cells of which are very much flattened out, go in deeper towards the median line to meet with its counterpart on the opposite side. The second tubule (figs. 92 and 93, pt.2) has become weaker, as a comparison of these figures with figs. 77 and 79 will show. On the contrary, the tubule of the next pair (fig. 94, pt.3) has much clongated and is bent considerably in dorso-lateral direction, so that we can no longer observe the nephrostome together with the tubule itself on the same section. The following tubule, the fourth (fig. 95, pt.4), is likewise well developed; the fifth (fig. 96, pt.5) is more or less weak in development as compared with the tubules of the two foregoing pairs. In short, these three pairs (from the third to the fifth) make parallel progress with the development of other structures, for instance, the mesenterial fold or the muscle-segments. This is a fact that is to be observed too in the younger embryo of this stage, as above described. At the present stage, we can find no trace of the tubules beneath the ninth myotome, where the tubules of the sixth pair ought to be found, but only the cross-sections of the collecting duct or the anteriormost part of the segmental duct are seen.

352 s. hatta :

Thus, the tubules of the third, fourth, and fifth pairs continue to grow, while the first pair has disappeared in the early part of the present stage (or at the end of the foregoing stage); the sixth has already commenced retrogression and the second is also growing weaker and weaker.

In the oldest embryo of this stage, there is to be seen no marked change in the pronephros, but the peritoneal lining is reduced into a very thin plate of a definite epithelium everywhere except at the pericardial portion, where the cells still have a columnar shape. The mesenchymatous cells accumulated on the median ventral line of the body are arranged in a certain order to be transformed into the cardiac tube. The third, fourth, and fifth tubules are also markedly prolonged and project into the body-cavity so as to cause the parietal layer of the peritoneum to fold between the epiblast and the body of the tubule (see this Journal: vol. x, Pt. xviii, figs. 8, 9, and 10). In some of the embryos, the tubules of the second pair undergo degeneration. I have met with, in this series of sections, the same condition of the sixth tubule as on the right of fig. 86, the right tubule having entirely disappeared.

#### Period 4.

The embryos in the next advanced stage (Stage v) are much diminished in size, assuming a form of a retort or of a pistol ('97, fig. 1, F). Figs. 98-106 represent sections through an embryo of this stage. The posterior larger section of the foregut comprising the pronephric region, has been reduced into a slender tube (fg.) which is bounded by almost a single layer of high cylindrical cells. The parietal layer of the peritoneum as

well as the coelomic projection (r.m.) is very much decreased in thickness and encloses the peritoneal cavity (pp.c.) that has now become spacious, while the visceral peritoneum is still thicker than other parts. The mesenchymatous cells found in the foregoing stage on the median ventral line are transformed into a thin layer of the endocardium of the heart and its anterior continuation (h. and tr.a.) which are suspended by the dorsal and the ventral mensenteries, and enclosed in the thick pericardial coat still forming a continuation of the peritoneum. I can detect, however, none of the traces of the peritoneal partition which was developed so markedly in the last stage that one could not possibly overlook them. I have endeavoured to trace the mode of disappearance of this structure, but have only found that in one lot of embryos the whole set of the structure was present while in the other no trace of it was perceptible. Unfortunately I have found no embryo in an intermediate condition.

The few cells observed from the last stage beneath the chorda, and also in the space outside the pronephric tubules on both sides are more or less multiplied. As the former group is transformed finally into the dorsal aorta, and the latter into the anterior cardinal vein of either side, I shall call them the tract of the dorsal aorta and of the anterior cardinal veins respectively.

Fig. 98 represents the section through the hind border of the branchial region. On either side of the enteric tube a small space (pp.c.) of the body-cavity is surrounded by the peritoneal epithelium, still consisting, in this part, of somewhat cubical cells. The ventral edges of the peritoneal membrane of both sides are just meeting at the median ventral line. A few mesenchymatous cells (tr.a.) found in the space between this meeting point and the ventral wall of the enteric canal, are destined to

form the anterior continuation of the cardiac tube or the truncus arteriosus. An irregular cell-structure (x) is seen on either side above the dorsal corner of the body-cavity and inside the tract of the anterior cardinal vein. It is this structure about which I could not at first decide with certainty whether it was a slice of the hind wall of the branchial chamber or a part of the pronephric tubule. All the cases examined, however, point towards its being a part of the tubule; the structure is detected in the anteriormost part of the body-cavity which wedges in, at about this stage, to the branchial region with a sharp angle (see fig. 97). The narrow space (fig. 98, pp.c.) found intervening between the structure and the peritoneal walls is a part of this eavity. One might suppose that the space may be the coelomic cavity of the branchial region; but, the space between the parietal and the visceral peritoneum of the branchial region is consolidated already in the preceding stage, being filled up with variously shaped cells of mesenchymatous nature (see fig. 97).

In the next following section shown in fig. 99, a tubular structure (pt.2) with an oval outline is seen on either side at the place where the pronephric tubule ought to be found. Its long axis is directed just like a tubule (compare with figs. 101, 102, &c.). This is doubtless a part of a pronephric tubule; but the corresponding nephrostome which ought to be found either in the section in front (fig. 98) or behind (fig. 100), can not be detected in either of them. The nephrostome must, therefore, be looked upon as having degenerated; and since this pair of the tubules is, in fact, detected underneath the fifth myotome, it must be identified as the second pair of the tubules. The section represented in fig. 100 shows on both sides the cross-sections of the collecting duct (cd.). On the left side, a cellular structure connects the

peritoneum and the collecting duct; it is the posterior wall of the tubule in figs. 99. Fig. 101 represents the section through the axial plane of the third tubule, the nephrostomes of which are recognized more clearly in the section behind it (fig. 102, nst.3). The tubules of this pair are comparatively not long. The fourth pair of the tubules and their nephrostomes are obvious in fig. 104 (pt. 4 and nst.4) which represents the third section behind that of fig. 102; the tubules much resemble those of the pair in front, showing the same convolutions as these. It is a peculiarity of the present stage that the aperture of the nephrostomes of the third and the fourth pair is not so wide open as in the last stage or as in more advanced stages! It is always nearly closed and slit-like, so that we can hardly trace the communication between the lumen of the tubule and the body-cavity.

Fig. 103 represents the section intervening between the sections shown in figs. 102 and 104. On the right side, the collecting duct alone, and on the left side, the duct together with a small part of the fourth tubule, is shown. The peritoneal membrane on the dorsal end of the body-cavity is folded far into that cavity (fig. 103, bs.). This fold is traceable from the anterior part of the third tubule to the hind part of the fourth (figs. 100-104, bs.). The space enclosed in this fold communicates freely with both the tract of the dorsal aorta under the chorda and the tract of the anterior cardinal vein outside of the pronephros and contains a number of mesenchymatous cells which probably wander in from the tract of the aorta and the anterior cardinal vein. As subsequent history shows, this structure constitutes the beginning of the glomerulus of the pronephros.

Figs. 105 and 106 represent two contiguous sections immediately posterior to the section shown in fig. 104. In fig. 105 we

356 s. hatta :

observe on either side the cross-section of the collecting duct (cd.) together with a part of the fourth tubule (pt.4); the longitudinal section of the fifth tubule (pt.5) is seen on the right side of fig. 106, standing in wide communication (nst.5) with the body cavity. This is the hindmost tubule. In the sections lying behind this, the cross-section of only the segmental duct is repeated.

Thus the tubules of the second pair undergo, at the present stage, complete degeneration. This process begins, in this case, as above seen, at the nephrostome and proceeds upwards to the collecting duct,—a process which is just the reverse of what is observed in the reduction of the tubules of the sixth pair and probably also of the first pair, in both which cases the tubules are first cut off from the collecting duct and the separation from the peritoneal cavity follows afterwards.

### Period 5.

In the Stage vt, embryos have developed so far that all the organs have received their definite forms and proper position with the exception of the middle and the hind portion of the gut, whose development is much delayed on account of the yolk-mass. Having absorbed the yolk-granules, the component cells of most organs are much diminished in size.

Figs. 107-110 have been drawn from a series of sections through an embryo in this stage. The enteric canal (fg.) is much diminished in diameter, presenting, in section, an elongated heart shape. The peritoneum becomes very thin in all its parts with the exception of the pericardium and the coat of the truncus arteriosus, in which its component cells are of cylindrical or cubical shape.

The peritoneal membrane lining the enteric canal immediately behind the branchial region is also thicker as compared with other parts (fig. 107), being composed of a single layer of cubical cells,—a peculiarity observed since the last stage (compare figs. 98-99 with fig. 107).

The pronephric tubules as well as the collecting duct are composed of a regular epithelium of cylindrical cells; the former, moreover, are much prolonged and, in some parts (fig. 108), much coiled, so that the peritoneal cavity which was almost a hollow space in the last stage, is filled up with the tubules and the cardiac tube.

Fig. 107 represents the section through the hind part of the sixth myotome; a pair of the tubules (pt.3) is hanging down in the body-cavity immediately behind the hind wall of the branchial On the right side, the axial plane of the tubule is chamber. cut through, while, on the left, the anterior wall of it is sliced; these are the tubules of the third pair. They show no bending in the antero-posterior direction, but are curved laterally and ventrally. The component cells are, in the nephrostomal portion, taller in comparison with those in other parts of the tubule or the collecting duct. The fourth tubule and nephrostome are seen on the right side of fig. 108, while on its left side, the communication of the corresponding tubule on the opposite side with the collecting duct is recognizable. The left nephrostome is found in the third section behind this, which is not figured. This pair of the tubules exhibits, in section, constrictions at two or three points owing to their curving somewhat in the antero-posterior direction (see the tubule on the right side of fig. 108). Fig. 109 is the section immediately behind the last and shows the cross-sections of the collecting duct (cd.) and a piece of the left fourth tubule (pt.4).

358 S. HATTA:

A pair of the glomeruli (figs. 108 and 109, ql.). is seen adhering on the median side of the tubule on each side and lined with the visceral peritoneum. The glomerulus represented in the last stage by a folding of the peritoneum which covers the tubules from the third pair to the fourth<sup>1)</sup>, is reduced, at present, into a pair of sacs of this membrane projecting on each side between the fourth and the fifth tubules; the other part of the folded membrane becomes adhered firmly to the walls of either the tubules or the body-cavity leaving no space of sacculation,-in short, a pair of long folds, extending from the anterior part of the third tubule to the fifth in the last stage, is reduced into a pair of sacs found in the position just mentioned. The inside of the sacs is compactly filled up with free-cells and communicates with the aorta tract and with the space outside the pronephros, where free-cells to be afterwards transformed into the anterior cardinal vein have been observed already from the foregoing stage.

The section represented in fig. 110 fortunately passes symmetrically through a pair of the nephrostomes (nst.5) and of the tubules (pt.5) hanging down in the peritoneal cavity. This is the fifth or the hindmost pair of the pronephric tubules in the present stage. The communication of the tubules with the collecting duct is seen in the section behind this. The tubules present also some antero-posterior bendings. Posterior to this, no tubule is found.

The pronephric tubules in the present stage are, therefore, reduced into the minimum number, i.e., three pairs<sup>2</sup>, all of which are retained so long as the organ functions as the excretory apparatus

<sup>1)</sup> See p. 355.

<sup>2)</sup> We occasionally find the four tubules to persist, and the additional tubule is the sixth-

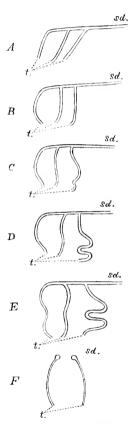
during the larval life of Petronyzon. Especially it must be noticed that the foremost pair of the persistent tubules (the third pair) is in close contact with the hind border of the hind wall of the branchial chamber where, in the foregoing stage, the second pair of the tubules was found, this latter having disappeared in the course of the last stage. It follows that the two somites, to which the first and the second pair of the tubules have belonged, have now entered into the formation of the branchial region.

The development of the pronephros after this consists only in the prolongation and the convolution of the tubules, no further change taking place with reference to the number of the tubules or to their histological structure, until the system undergoes degeneration to be replaced by the mesonephros, which functions as the excretory organ for the whole subsequent life of *Petromyzon*.

The convolution of the tubules is hard to make out. I have reconstructed them from a number of sections; some of these are diagramatically given in the annexed woodcut (Woodcut 2).

With the growth of the muscle-segments the collecting duct is prolonged, so that the points of connection of the tubules with that duct become farther apart from one another, while the nephrostomal portions of the tubules retain more or less their original positions; in this wise, the tubules are laid in oblique positions directed anteriorly and posteriorly (A) and have no other curvature than the ventro-lateral bending (the frontal projection of the curvature is shown in F). Then the antero-posterior bending begins to take place. The foremost tubule is curved forwards in its whole length, while a small curvature in the distal

(nephrostomal) portion of the two following pairs is directed backwards. The nephrostomes retain their first position (B).



Wood-cut 2.—Diagrams showing the convolutions of the tubules in later stages. t. pronephric tubules. s.t. segmental duct.

Now the secondary curvatures take place (C). The nephrostomal part of the foremost pair is crooked just like that of the two hind pairs in B; the middle tubule is bent forwards like the foremost tubule. The hindmost tubule makes a small forward curvature and a large backward bending. In the next stage, D, the foremost and the middle undergo no marked change, but the secondary curvatures of the hindmost tubule are much more strongly expressed. In E the foremost receives a secondary curvature directed backwards at the middle part; the middle acquires a curvature in opposite direction; the hind tubule undergoes no marked change except in the increased degree of the original curvatures. It seems that the subsequent bendings always take

place in the curved portion until there arises a system of complexly convoluted tubules filling up the chest cavity.

As has been said, throughout these phases the positions of the nephrostomes are not markedly changed, retaining the same condition for a considerable period. The bendings of the tubules are caused, therefore, by the growth of the tubule at the point of bending.

The curvature in the ventro-lateral direction is very simple and undergoes no remarkable change; its projection is shown in F.

# B.—The Segmental Duct and the Genital Cells.

For the sake of simplicity, the development of the segmental duct and of the vascular system in the pronephros has been entirely put aside in the description given above.

As already alluded to, the origin of the segmental duct in Petromyzon is extremely difficult to make out, because its formation goes on rapidly at a comparatively young stage. The early process of its formation is essentially the same as in the pronephric tubules. In the anterior region, the intermediate cell-mass or the nephrotome (see p. 340) behaves itself in precisely the same manner as in the Anlage of the pronephric tubules; the difference is that it is cut off from the lateral plate and is transformed into the duct, while in the case of the tubule it retains the continuity with the lateral plate. If fig. 31, which represents the section through the tenth somite (i.e. the somite, from which backwards the Anlagen are converted to the segmental duct) be compared with the left half of figs. 2, 5, 6, 14, and the right half of fig. 16, in which the Anlagen all develop to the pronephric tubules, it will be found that there is no difference between them; in fact, they are morphologically equivalent to one another. Such an Anlage is, posterior to the pronephros, not confined to the tenth somite, but, as has been already repeatedly said (pp. 317, 320, and 327), is observed for some segments further backwards (see fig. 17).

The Anlage thus pronounced in each somite soon assumes a characteristic oval form, being completely cut off from the myotome to which it belongs (compare the right side of fig. 3 with the right side of fig. 58 and see the description on p. 335). The mode of constriction is also the same as in the case of the pronephric tubules; the indentation begins at the anterior and posterior borders of the somite, and the middle portion is cut off last (compare with the explanation on p. 320).

Here also, the coelomic projection is formed in the same mode and at the same point as in the case of the pronephric tubules (see left side of fig. 63, c.p.).

Up to about this time, the Anlage shows a feature much resembling that of the tubule, so that one who has not followed its further history might mistake it for a pronephric tubule (compare the left side of fig. 63 with figs. 67-74). But cell-mutiplication which occurs almost invariably in the case of the pronephric tubules, is not observed in the Anlage of the segmental duct which is soon cut off from the lateral plate (including the coelomic projection) and assumes a characteristic tubular structure composed, in cross-section, of radially arranged cells of columnar shape. Its position is always on the parietal aspect of the dorsal (proximal) angle of the peritoneum where the coelomic projection passes over into the lateral plate (see fig. 75). This separation of the Anlage of the duct from the lateral plate goes on, it seems to me, on the whole from the anterior part to the posterior, but often irregularly; for not infrequently, the duct in some anterior somite is connected with the lateral plate, while it is already cut off completely in posterior somites. In fact, there are some somites in which the separation is very much delayed and I have often been surprised to find what appeared like a pronephric tubule in a

somite (see fig. 63) far backward of the posteriormost tubule which is found in the ninth somite.

The segmentally arranged Anlagen of the segmental duct are secondarily united with one another just as in the case of the collecting duct in the pronephric region. This union seems to take place during the separation of the Anlage from the myotome and is finished before it is separated from the lateral plate; for, when the Anlage first comes into view, there is no intersomitic cord as in the case of the pronephric tubules and the duct is seen already consisting of radially arranged cells (fig. 58) when it is cut off from the lateral plate. When established, the duct is the same in structure in both the somitic and intersomitic spaces; a cross-section of such a duct in the intersomitic portion is shown on the right side of fig. 75, while that in the somitic portion is seen on the left of the same figure.

This condition of the duct is already traceable, in Stage III, for no fewer than 10 somites from the hindmost pronephric tubule backwards, and it forms a direct posterior continuation of the collecting duct. The duct remains awhile as a solid cord of cells arranged radially in cross-section, but it soon acquires a lumen (figs. 75, 86, and 87, sd.). The further development of the duct goes on more promptly than that of the tubules in the hind part, and therefore, the embryos at such a stage (Stage III) have a well developed duct and more or less primitive tubules (compare fig. 74 with fig. 75).

In the hind region, where yolk-cells are crowded, the process is much delayed and more or less modified. Instead of the differentiation of the cells in situ, it seems to me, a few cells are detached from the nephrotome; a number of cells is produced by repeated division of these cells (fig. 19, a.sd.) and becomes ar-

364 s. hatta :

ranged as in the Anlagen in the anterior region. Fig. 89 represents the section through the twenty-eighth somite in the series of sections shown in figs. 77-86; it is the hindmost section in this series of sections, in which the cells just spoken of are detected; there are found a few cells (a.sd.) of this kind which show no definite structure, but are scattered. In the next anterior section (fig. 88) the cells are arranged more or less radially. In the sections lying further anteriorly to this a perfect tube is formed as seen in fig. 87 (sd.) which shows the frontal section through the seventeenth to twenty-third somites in the same series as the above two figures.<sup>1)</sup> In what somite this modified mode of the formation of the duct begins I can not tell with exactness, but it is certain that the duct arises by the differentiation of the nephrotomic cells in situ more than 10 segments back of the hindmost pronephric tubule. I have considered it possible that these cells (u.sd.) might be epiblastic in origin, but I can not find that the cells composing the epiblast over this cell-group show any sign of mutiplication; while on the other hand, the cells on the dorsal edge of the lateral plate (which corresponds to the nephrotome in the anterior part) are very active. I see, therefore, no escape from the conclusion that these cells are mesoblastic in origin.

Also in the anterior part of the body, the epiblast consists throughout these phases of development always of a single layer of cubical cells and shows a sharp contour against the structure inside it, being, in most cases, intervened by a space. Naturally, mitotic figures are observed at several points, but the products of these cell-divisions contribute only to the extension of the epiblast itself, as may be inferred from the direction of

<sup>1)</sup> By the bending of the body-axis, some sections in a series of cross-sections are unavoidably cut through frontally.

the spindles, the long axes of which are directed always parallel to the surface of the layer. I have nowhere observed any trace of either the proliferation or of the easting off of cells from the epiblast to give rise to the segmental duct.

In the cloacal region, the formation of the segmental duct goes on a little earlier than in the region next anterior to it. In spite of much effort, I failed to observe the very beginning of the formation at the cloacal opening, and I have nothing to tell of its earliest stage. In the series of sections from which figs. 77-89, are drawn, I can not yet find in the adjacent part of the cloaca any trace of the duct; but in the section represented in fig. 90 which passes through the dorsal lip of the blastopore of an embryo with about the same number of the mesoblastic somites (34 or 35) as the one just referred to, the duct already breaks through into the cloacal cavity (co.sd.)<sup>11</sup>. Fig. 91 represents the next ventral section which passes through the dorsal part of the blastopore (bp.). As seen in these two sections, immediately inside of the blatlopore (bp.), where the hypoblast passes over into the epiblast, the walls of the cloacal cavity send out, right and left, a symmetrical pair of diverticula<sup>2</sup> (c.dv.), forming an acute angle, the inner side of which is a part of the enteric wall, while its outer side is the direct continuation of the epiblast. The walls of this diverticulum pass over into the segmental duct (sd.). The communication of the segmental duct with the cloacal cavity is found, therefore, at the point where the epiblastic layer of the lip is reflected inside and passes over into the hypoblast. This point of communication is, however, shifted far inside and

<sup>1)</sup> This opening is found in the same vertical plane as the 34th or 35th somite.

<sup>2)</sup> The right diverticulum only is seen in figs, 90 and 91, the left one being observed in another section which is untigured.

dorsally when the development proceeds further (fig. 111, co.sd. and c.dv.).

I have also met with two cases (figs. 90 and 111), in which I have observed some epiblastic cells of the external lateral walls of the blastopore multiplying actively and having mitotic spindles (x) with axes directed perpendicularly to the plane of the epiblast, while the duct comes in firm connection with that point of the epiblast,—the connection is so firm that the duct and the epiblast appear to form one and the same tissue. At this point, thus, there is every appearance of epiblastic cells partaking in the construction of the segmental duct.

The collecting duct pertaining to the ninth somite forms the segmental duct in that segment, having lost the connection with the tubule.

Up to Stage II, the duct is represented by the segmental Anlagen in about 8 segments back of the ninth somite; in Stage III, these Anlagen are converted into the duct in about 10 anterior segments; while in the course of Stage IV it opens out into the closeal cavity.

From the above account, it is easily conceivable that the Anlage of the segmental duct and that of the pronephric tubule are perfectly homologous, and that the duct is a continuation of a series of abortive pronephric tubules in the hind region.

Underneath ten and more myotomes lying posterior to about the fifteenth somite the proximal portion of the lateral plate, which corresponds to the nephrotome, contains peculiar large cells (figs. 87, 88, and 89, gc.) loaded with an enormous quantity of yolk-granules; the other mesoblastic cells in this part, being much flattened out, form a thin layer over these cells. These peculiar cells are, I think, the equivalent of the primitive genital cells found in the corresponding part of the Amphibian and Selachian body.

Up to Stage III, these cells can not be distinguished from other mesoblastic cells which are equally rich in yolk-granules. In Stage IV, they become conspicuous; and in Stage V, again indistinguishable from other constituent cells of this part.

# C.—The Vascular System in the Pronephros.

In early stages, no trace of the vascular system is perceived in the pronephros. What is recognisable as a fore-runner of the vessel is represented by mesenchymatous cells scattered in the space between the primary germinal layers (figs. 77 and 82, mch.). These free cells are detected, during Stage IV, in three tracts, viz., beneath the chorda, beneath the ventral wall of the enteric canal and outside the pronephric tubules on either side (figs. 77, 79, 80, 81, and 82, mch.). In Stage v, or at the end of Stage IV, the cells below the hind section of the fore-gut are converted into the endothelium of the heart and of the vessels which are its direct continuations. The cells beneath the chorda are destined to be transformed into the dorsal aorta, and the cells on either side of the pronephros constitute the first indication of the cardinal veins. It is these three vessels—the aorta and the two cardinal veins—which come in relation with the pronephros.

In the embryos in which the degeneration of the tubules is still going on, there is no special vessel supplying the pronephros; but when the process is over, a pair of long blood-spaces (figs. 100-104, bs.) is found in communication with the aorta-tract.

<sup>1)</sup> A few of them are observed here and there already in Stage III.

They are the spaces formed by the slackening and folding of the median peritoneum which coats the pronephric tubules, as above stated (p. 355). The fold, i.e. the space, extends throughout almost the whole length of the pronephros (figs. 100-104) and contains numerous free-cells. But, as the peritoneum finally adheres to the median walls of the tubules in each nephric segment, the space becomes divided into three pairs: the anterior pair, which soon disappears, is found between the tubules of the second and third, the middle is detected between the third and fourth, and the posterior between the fourth and fifth tubules. These spaces communicate directly,—medially with the aorta tract and externally with the tracts of the anterior cardinal veins, which emerge, in later stages, in the pronephros. They are the blood-spaces which, I believe, correspond with the intersomitic arteries demonstrated by PAUL MAYER and others in Selachia.

When the tubules develop further, the arterial portion of these blood-spaces disappear except the middle portion where it is sacculated and filled up with the mesenchymatous cells (figs. 108-109, yl.). This portion is the structure which is called the glomerulus of the pronephros; it is found one on each side (see pp. 355 and 358).

Having followed, in the foregoing pages, the successive processes which take place in the development of the pronephros in *Petronyzon*, step by step, I will give a short resumé of the facts.

1. In the earliest part of Stage II, the mesoblast consists simply of [the parietal (dorsal) and the visceral (median and ventral) layers. The proximal portion of the mesoblast is dis-

tinguished, first of all, in the histological structure from the distal portion: the former is composed of columnar cells, and the latter of irregularly shaped cells. Only the proximal portion which occupies the largest part of the mesoblast undergoes the metameric segmentation and gives rise to the scleromyotome and the nephrotome (in the sense of Rückert); the distal smaller portion remains unsegmented and is later converted into the flattened epithelium of the peritoneum.

- 2. The earliest traces of the pronephros are noticeable in exceedingly young stages, that is, in the early part of Stage II, in which the embryo has about 16 somites.
- 3. They are expressed in the form of a diverticulum of the parietal layer of that section in each mesoblastic somite, which forms the ventral half of the segmental part of the mesoblast and is called the nephrotome. This is the Anlage of the pronephric tubule and not of the segmental duct.
- 4. The pronephric diverticulum or the Anlage is brought about by the evagination of the parietal layer in each nephrotome, enclosing a part of the primary coelomic cavity.
- 5. The nephrotome is separated from the proximal portion of the segmented mesoblast and forms awhile the proximal portion of the unsegmented mesoblast or the lateral plate. The separation begins with an indentation in the anterior and posterior borders of the mesoblastic somite; the myocoelome communicates for some time by a narrow passage with the general coelomic cavity.
- 6. The Anlage has no histological connection either with the preceding or the following Anlage or with the other germinal layers; it is, therefore, segmental in origin and myomeric in position.
  - 7. The Anlagen are developed, in Stage II, in about 12

segments and are cut off from the scleromyotome in 4 segments. In Stage III, the separation of the Anlage from the myotome goes as far backwards as the sixteenth or seventeenth segment.

- 8. The anteriormost Anlage is found in the hind part of the fourth somite and is the first to arise; the second follows it, and so forth.
- 9. The Anlagen in each somite are secondarily united with one another by the solid cellular cord which is budded out of the anterior and posterior rims of the Anlagen themselves; thus the collecting duct (Sammelrohr in the sense of Rückert) is established. This process is originally to be looked upon as the coming together of the ends of the tubules.
- 10. The canalization of the collecting duct begins within each Anlage and proceeds, generally speaking, posteriorly, until the Anlagen in front and back are put in free communication.
- 11. Each Anlage grows dorso-laterally and acquires a tubular form. The collecting duct is shifted gradually in a dorso-median direction; finally it comes to lie between the myotome, the mesentery, and the *chorda dorsalis*.
- 12. The tubules open in the coclomic cavity at the lateral angle of the dorsal corner of that cavity.
- 13. In the somites posterior to the ninth, the tubules are, during Stage III, cut off also from the lateral plate and establish a long duet running, on each side, along the dorsal aspect of the lateral plate where originally the tubules opened. This is the segmental duct; the tubules and the collecting duct in the somites anterior to this constitute the glandular part of the pronephros.
- 14. The glandular part, or the pronephros proper comprises six somites, from the fourth to the ninth. The maximum number

of the pronephic tubules which is attained by the embryo in Stage III, is, therefore, six pairs.

- 15. The tubules of the first and second pairs come, in Stage III, temporarily in close contact with the epiblast, but do not receive cells from it; they soon return to their original condition.
- 16. The anterior extremity of the system shows, from the first, degenerating features. The first, second, and sixth of the tubules begin, during Stage III, to decline; and at the end of Stage IV, or the beginning of Stage V, the tubules are reduced into the minimum number, which consists of three pairs from the third to the fifth. These three pairs function as the actual excretory organ for a considerable length of time.
- 17. Retrogression is first met with in the first pair of the tubules, which decline probably without further development, soon after their separation from the myotome is completed; they seem to atrophy from the free end. The next pair degenerating is the sixth, which is at first cut off from the collecting duct and remains for a short time, but soon disappears without leaving a trace. The second pair persists for some time seemingly to function as the excretory organ, but it atrophies already in the early part of Stage v, the communication with the coelomic cavity being first obliterated; and in Stage vi, none of the structure remains to be recognized.
- 18. The foremost pair of the persistent tubules comes to lie in close contact with the hind wall of the branchial chamber. The two mesoblastic somites which correspond to the first and second nephromeres should therefore be looked upon as having entered into the formation of the branchial region.

The stages in which the tubules appear and abort in different somites are shown in the annexed table.

	Yem. 1	!	Som. 11	Som. III	Som, IV	Som. V	Som, VI	Som. VII	Som. VIII	Sem. IX	Som. N	Som. XI	Som. XII
Stage H [					Ani. I	Aul. 2	Λnl. 3	Anl. 4	Anl. 5	$\Lambda$ nl. 6	Anl. 7	Anl. 8	Anl. 9
Stage III					Tub.1	Tub.2	Tub.3	Tub.4	Tub.5	Tub.6	Segu	nental (	duet.
Stage IV						Tub.2	Tub.3	Tub.4	Tub.5	Tub.6	Segn	nental	duct.
Stage V						Tub.2	 Tub.3	Tub.4	Tub.5	2	egment	al duci	
Stage VI							Tub.3	Tab.4	Tub.5	S	- Segm <b>e</b> nt	al duc	1.

- 19. In older embryos of Stage III, the visceral layer of the nephrotome is folded out, and is called the coelomic projection which resembles the coelomic pocket described by Price for Bdellostoma; however, in Bdellostoma, the fold is derived from the parietal and visceral layers of the lateral plate and is afterwards converted into the Bowman's capsule, whereas the coelomic projection is the product of only the risceral layer of the nephrotome; it gives rise to the radix of the mesentery which offers materials to the mesonephric tubules and to the gonads.
- 20. The topographical position of the pronephros becomes first definite in Stage IV. It is situated in the chest cavity, dorso-lateral to the heart, forward of and dorsal to the liver, extending along either side of the chorda. This position is somewhat changed as the development proceeds; the pronephros comes, in later stages, in front of the liver.
- 21. During Stage IV, a structure, which I have called above the *peritoneal partition*, is observed as an outgrowth of the peritoneal wall and disappears during the same stage.

This horizontal partition is found at three levels. The most dorsal is well developed, the ventral is a mere trace, and the middle is intermediate between the above two. I can not state at present anything definite as regards the significance of this structure.

- 22. The convolution of the pronephric tubule takes place in Stage IV. With the growth of the myotome, the collecting duct is prolonged; consequently the connecting points of the tubules with the duct are farther removed from one another than before, whilst the nephrostomes retain their original position; so that, the two posterior pairs of the tubules are placed in an oblique direction from dorsal and caudal to ventral and cranial. Each tubule is, then, convoluted in a cranio-caudal direction between the heart and the lateral peritoneal wall. In older stages, the tubules are coiled in all directions, until the chest cavity becomes filled up with the convolution of the tubules.
- 23. Up to Stage vi, the nephromeres and the myomeres exactly coincide one above the other in position. This period is very long in comparison with other Craniota. As the development proceeds further, the pronephic tubules are however shifted gradually backward, so that, in Ammocoetes 10 mm. long, the myotomes are already not situated upon the tubules pertaining to each of them. In later stages, nothing of the relation can be traced.
- 24. The segmental duct is looked upon as being brought about by the union of a series of the abortive pronephric tubules in about 12 somites lying posterior to the eighth somite. The Anlage is laid in the parietal layer of the nephrotome in exactly the same manner as in the pronephric tubules of the glandular part.

<sup>1)</sup> In the 9th somite, the ab rted tabule actually forms the duct in the segment.

The difference is, that the tubules in the posterior region are soon cut off from the lateral plate and become the duct.

- 25. Between the epiblast on one hand, and the Anlage or the duct on the other, there exists always a space, and the duct has no connection with the epiblast except at its posteriormost end where the epiblastic cells might, as judged from the mitotic figures, contribute to the formation of the duct.
- 26. In the somites posterior to about the twentieth somite, the Anlage of the duct is represented by a few cells in each segment probably detached from the dorso-lateral angle of the nephrotome. These cells multiply and are transformed into the segmental duct in the posterior part.
- 27. In Stage II, the Anlagen of the segmental duct are cut off from the mother layer in a few somites; in Stage III, the duct is formed as far as about the eighteenth somite, while in Stage IV, it breaks out into the cloacal cavity. The cloacal opening of the segmental duct is found at a point where the hypoblastic cloacal wall is reflected into the epiblast, these two layers forming a diverticulum on either side.
- 28. In stage IV, the primitive genital cells become apparent in the nephrotomes of the posterior 10 or more somites; they can not be discriminated from other mesoblastic cells in the next advanced Stage.
- 29. The blood-vessels, which specially supply the pronephros, acquire definite form in comparatively later stages, viz., at about Stage v. The dorsal agree pours out the blood into two pairs of the blind vessicles, which are formed by the folding of the parietal peritoneum and are found between the first and second pairs, and between the second and third pairs, of the persistent tubules

respectively. The venous blood is carried away through the anterior cardinal veins which penetrate the pronephros.

29. These blood-spaces are thus segmental in arrangement and intersomitic in position. The two anterior pairs of them soon undergo atrophy, but the posteriormost pair persists, becoming enlarged and sacculated at the distal extremity. This sacculated part of the vessel is filled up with free-cells and is called the glomerulus of the pronephros, and, therefore, there is only a pair of glomeruli in *Petromyzon*.

## II. Historical Review and Conclusions.

As is well known, Max Schultze ('56) was the first who discovered the pronephros in *Petromyzon*. Having investigated the larvæ of *P. plancri*, the author describes the structure as "Drüsenanlage" and homologised it with the "Urnieren (Wolf'sche Körper)" of the frog's larva. His statements on this body are as follows: "Nicht lange nach der Bildung dieser Drüse (Thymus) entsteht die Anlage einer zweiten, aus dem unter der Chorda dorsalis angehäuften Blastem über dem Herzen. Aus der durch Pigmentanlagelungen früh schon sehr undurchsichtig werdenden Masse wachsen nämlich nach unten, gegen das Herz zu, 3 oder 4 kurze Fortsätze hervor, welche eine eigenthümliche Wimperung zeigen" (p. 30).

The stage spoken of probably corresponds to Stage v, or vi, of my embryo.

Our knowledge on this subject received important additions by the noted investigations of W. Müller and Max Für-Bringer. Muller ('75) noticed the first traces of the pronephros in a very young embryo, which had yet only four pairs of gill-slits. This Anlage gives rise to a much coiled gland, which opens into the body-cavity, at first through only one ciliated funnel, but afterwards through four. The gland passes over posteriorly to a pair of ducts, which run along the chorda on either side and open into the cloaca. Müller has homologised the structure with the "Vorniere" of Myxine and called the duct "Urnierengang" (pp. 121-122). He found a pair of glomeruli projected on the median surface of the gland and lined with the peritoneal epithelium.

Max Fürbringer ('78) studied the larvæ of Ammococtes planeri, which varied from 4.5 to 180 mm. in length. His statements essentially confirm Muller's. In his account we find the following sentences: "Die auf allen Präparaten ausgebildete Vorniere, die ich im Wesentlichen ganz wie Müller fand, bildet einen nahmentlich bei den mittleren Stadien voluminösen und durch 4-5 Myokommata erstreckten Complex von Windungen, die vorn durch mehreren Peritonealcanäle (Wimpertrichter) in Bauchhöhle münden und hinten in den Vornierengang übergehen. Diese auf die 2-3 ersten Myokommata beschränkten Trichter ragen in unregelmässiger Folge bald ventral-medial, bald ventrallateral in die Bauchhöhle vor und wurden (von Carlberla und mir) meist zu fünf gefunden. Die von rundliche Epithelzellen bekleidete Glomerulus verhielt ganz wie Müller beschreibt" (p. 42).

The larvæ of Ammocoetes in question seems to correspond probably to Stage v, or later stages of my list; in such a stage, I could not find more than three (or rarely four) pairs of the tubules, or of the nephrostomes.<sup>1)</sup>

<sup>1)</sup> See the foot-note on p. 358.

The authors who have investigated the development of Petromyzon embryos step by step, are W. Scott, Goette, Shipley, and v. Kupffer. Their opinions are, however, somewhat divergent. Scott ('82') derives the pronephric tubules from the segmental duct which is, according to him, brought about by the differentiation in situ of the cells forming the proximal margin of the lateral plate. The process takes place in the whole extent at the same time. At certain points (segmental?) of the duct thus formed, evaginations are produced out of it; these evaginations subsequently open into the body-cavity and establish the nephrostomes which are, according to Scott, found from two to three pairs in number. At about the stage in which the funnels are formed, he observed a pair of glomeruli.

"In most respects," Shipley's observations ('87) "confirm his" (Scott's). But "on the origin of the ciliated funnels, the results differ from Scott's" and agree with those of Fürbringer (Amphibian pronephros?). According to Shipley, "in the region of the heart, where the body-cavity has already appeared, its origin (i.e., of the segmental duct) seems to be somewhat different. The lumen of the segmental duct here becomes continuous with a groove in the parietal peritoneum, lying near the angle where the somatopleure and the splanchnopleure diverge. When this groove closes it leaves four or five openings which persist as the openings of the ciliated funnels" (p. 20).

v. Kupffer<sup>1)</sup> ('88) observed, in *P. planeri*, the three pairs of the tubules arising from three distinct evaginations of the parietal

<sup>1)</sup> I know this paper only by the abstract in: Jahresbericht ü. die. Fortschr. d. Anat. u. Physiol., Bd. 17, 1889.

378 s. hatta:

layer of the lateral plate; the segmental duct is looked upon as of the epiblastic origin.

GOETTE ('90) worked the development of Petromyzon fluviatilis; his results with respect to the pronephros show some agreement with mine, especially those concerning the later stages. The author derives also the whole system of the pronephros (including the segmental duct) solely from the mesoblast. But we diverge in some important points from each other; he has found the earliest traces of the structure at a time when the rudiment of the heart first becomes apparent (his vi. Periode) (p. 64). From the account given in the foregoing pages it is clear that this period belongs to a later stage in which the pronephros has already made a considerable progress in development; his figures 99, 103, &c., which are spoken of as representing the first appearance of the structure, approximately correspond with my figures 82, 83, &c., and with those of even older stages.

The pronephros is, according to Goette, not of a separate Anlage in its first appearance, but arises in a form of a longitudinal furrow formed, on each side, by an evagination of the parietal layer of the mesoblast; the lips of the furrow being fused at certain points, there remain three openings; these are converted afterwards into three tubules and ciliated funnels. The tubules are added by stages until there are usually five, or more rarely four or six; but how these are multiplied, he can not say with certainty. The tubules have, it seems to him, no relation to the metameres of the body; for 3 to 5 tubules are found in the extent of 2 to 3 metameres (loc. cit., pp. 64-65).

The segmental duct originates, according to GOETTE, in precisely the same way as the pronephros proper; the only difference is the complete constriction of it from its mother-layer just as

I have made out. From the region of the liver-anlage backward the development of the duct is irregular; he says: "Auf der einen Seite zeigt sich seine Anlage noch rinnenförmig, während sie auf der andern Seite schon vollkommen röhrenformig abgeschnürt ist. Endlich wechselt dies Verhalten auch auf derselben Körperseite, so dass derselbe Gang, von der Lebergegend rückwärts verfolgt, bald rinnen-, bald röhrenförmig, geschlossen oder mit offener Lichtung sich darstellt" (loco cit., p. 56). The hind end of the duct opens in the cloaca (Afterdarm) by the fusion of their walls and by the communication of the lumen of the duct and the diverticulum of the cloaca. I have not observed in any stage of my embryos examined the numerous convolutions of the segmental duct demonstrated by Goette in the region immediately behind the "ursprüngliche Kopfniere."

Goette has made out the three "peritoneale Scheidewände," as he calls them: two respectively in the anterior and the posterior end of the pronephros, and the third on either side of the liver. Later, the first contributes, according to him, to the formation of the hind wall of the branchial pouch (Kiementasche); the second is converted into "eine Venenbrücke zwischen dem Sinus venosus and der Leibeswand," while the third disappears without leaving a trace. They are, according to Goette, homologous with the "Schlussplatte" of the pronephros in Teleostei; considered phylogenetically, nevertheless, they have no intimate relation to the pronephros in Petromyson (loco cit., pp. 56-61). This structure is, as stated on p. 349, doubtless the same as the uppermost peritoneal partition which I have found in my embryos. I have nothing to communicate on its significance; but I feel sure that his statement is not accurate when he says the structure appears earlier than the pronephros; for his figs.

380 s. hatta:

96 and 97, to which his statement refers, represent a stage considerably later than the first formation of the pronephros itself. And the peritoneal partition is not confined to these three points, but is continuous throughout the whole extent of the pronephros; moreover, beside the "peritoneale Scheidewände," there are found two other partitions of a similar character as above stated. Also, as to the fate of the structure my results differ from his: I have not been able to observe at all any such contribution to the formation of the hind wall of the branchial chamber and of the "Venenbrücke," as is affirmed by Goette.

Rable ('96) says in his recent extensive work on the Selachian nephric organ, that in quite young larvæ of *Petromyzon fluviatilis* the pronephros also begins in the seventh somite, in which the first of the four ostia are found, as in *Pristiurus*. His larvæ are, however, 501 hours or 20 days and 21 hours old; such larvæ correspond to my embryos in Stage vi, and upwards, in which anteriorly two pairs, and posteriorly, one pair of the tubules disappeared and only three persistent tubules are seen. His first nephrostome represents the foremost of the persistent nephrostome.

The accounts cited above all agree with the results given in the present paper in deriving both the pronephros and the segmental duct from the mesoblast alone, with the single exception of v. Kupffer who assumes the epiblastic origin of the segmental duct. They differ from the account given in the foregoing pages in the mode of the formation and in the number of the tubules formed. The first point of difference is due to the

<sup>1)</sup> See the reference under Selachia (p. 390).

fact that the authors probably overlooked the earliest phases of formation, which take place, as shown above, in a stage very young, but not younger in comparison than that in other Anamnia; for the formation follows the metameric segmentation of the mesoblast in the anterior region. In later stages, the tubules and the intersomitic portion of the collecting duct repeated in sections of a series appear, indeed, like the cross-sections of a longitudinal furrow or groove of the lateral plate, the lips of which are fused at certain points, as described by Shipley and Goette (see my figs. 66-74).

The number of the tubules and nephrostomes varies according to the stages of development. And if some stage or stages are overlooked, it must necessarily lead to an erroneous conclusion. This is the probable reason why the statements of the writers with reference to the number differ.

Indeed, the anterior extremity of the pronephros has already, from the first appearance, the features of a rudimentary organ; the first pair of the tubules can not be observed at the same time with the following five pairs, except by extremely good luck. In some embryos of Stage III, we see occasionally the collecting duct alone in front of the first tubule, so that we are led to infer that there were some pairs of tubules in front of the present first pair, which have degenerated during the course of the ancestral history.<sup>1)</sup>

As is seen above, all investigators who have been occupied with the study of the development of *Petromyzon* agree in describing only one pair of glomeruli. Shipley says "there is only one glomerulus on each side, stretching on each side of the

<sup>1)</sup> I have stated above that in the earliest part of Stage III, the anterior extremity of the left collecting duct presents a conical protuberance (see the footnote on p. 329).

alimentary canal extending through about the same space as the glandular part of the kidney. Each glomerulus is a diverticulum of the peritoneum, which generally becomes sacculated;......." (p. 21). The statements by Goette confirm Shipley's, and my results also agree with theirs. However, this is not all of the vascular system of the pronephros but represents a posterior portion of it, the anterior part having disappeared entirely (see p. 368).

No previous writer on *Petromyzon* has described such early stages as given above in the development of the pronephros, nor has any one remarked the temporary existence of the pronephric tubules in the branchial region as well as in the region of the segmental duct. I will, therefore, extend the comparison over the allied groups such as Myxinoids and *Amphioxus*, and higher Craniota to verify the new facts.

With reference to the development of the nephric organ in Myxinoids, there is a great deal of information which we owe to the unwearied labors of W. Müller, Semon, Weldon, and others. They had, however, no opportunity to observe the earliest stage of the embryos. Recently our knowledge on this subject has been greatly augmented by the new works of Price, Dean, and Maas.

Price ('97) worked out the early development of the pronephros observed in a few embryos at different stages of *Bdellostoma* stouti. According to him "the first indication of the system occurs here in the eleventh segment (of spinal ganglion), and consists of a simple thickening of the somatic layer of the coel-

<sup>1)</sup> I have not seen the paper by J. MÜLLER.

The pronephros in *Bdellostoma* comprises, according to Price, 69 segments (spinal ganglions). As it begins at the transverse plane opposite the eleventh spinal ganglion, it is inferred that the pronephros in *Bdellostoma* is extended over the whole length of the branchial region. But "the excretory system disappears through the greater part of this region before the gills are formed" (*loro cit.*, p. 217).

The segmental duct (in s. str.) is, according to the author, brought about by the rudiments of the hinder 20 degenerated tubules (in his Stage C); the number of the declining tubules increases by stages: in Stage A, there are two; in Stage B, nineteen; and in Stage C, twenty.

This account is thus in close agreement with that given in the present work, excepting a slight difference as to the origin of the collecting duct and as to the number of the tubules. In *Bdellostoma*, the collecting duct develops out of the Anlagen independent from that of the tubules, while in *Petromyzon*, as stated in the foregoing description, the Anlage in a mesoblastic 384 s. hatta:

somite develops solely into the tubule, and by the secondary union of the tubules' ends, the collecting duct is brought about.

As regards the number of the tubules, there are, in *Petromyzon*, only two pairs in the branchial region instead of twenty in *Bdellstoma*. The number is, however, of secondary importance; it varies with the stages of embryos and possibly with individuals, and naturally more with the embryos of different families. This numerical variation is readily explained by the degenerating tendency of the tubules.

Price has made out the segmental evaginations of the dorsal corner of the coelomic cavity corresponding to the nephromeres; they are called by him the "coelomic pockets." In Petromyzon, I have found a series of solid knobs on the visceral layer of the intermediate cell-mass, which are transformed into the segmental folds of epithelium, forming then the direct continuation of the peritoneum. Thus the coelomic pocket in Bdellostoma and the coelomic projection in Petromyzon are apparently very similar structures; the two, however, differ from each other in origin and in fate. The former (coelomic pocket) is constructed by the parietal and visceral layers of the lateral plate, while the latter (coelomic projection) is the product of only the visceral layer of the nephrotome, the ventral half of the segmented part of the mesoblast. The coelomic pockets become the Malpighian body, and the coelomic projections give origin to the radix of the mesentery, from which the gonad-cells and the mesonephric tubules are derived. Nevertheless, these two structures are, I believe, homologous. Price's statements on the derivation of the coclomic pocket from the two peritoneal layers, are not as clear as is desirable, and its partition from the body-cavity might, it seems to me, represent the uppermost peritoneal partition which soon disappears, in *Petromyzon*, without any definite significance. At any rate, the structure represents "parts of the original segmental coelome, that is, the nephrotome," an unmistakable fact which is denied by Price.

The embryos of Myrine which formed the materials of the valuable works by Maas are too old to be compared with those of Petromyzon used in the present work. But the results obtained by the author differ from those of Price in an important point, namely, in the derivation of the mesonephros.

The pronephros and mesonephros are, according to Price, different parts of the same organ. "If the organ in question could only be a pronephros alone, or mesonephros alone," says Price "I should unhesitatingly pronounce in favour of its being a pronephros" (loco cit., p. 120). And he proposes to call "the entire embryonic kidney holonephros." With Rabl, Maas, and others, I hesitate to accept Price's conclusion; for there are, as may be inferred from his statements, great gaps not only between the Stages B and C, but also between Stage C and the adult. The formation of the mesonephros takes place in Petromyzon only at a stage much advanced, in which the processes of the formation and degeneration of the pronephros go on in much the same manner as in Bdellostoma, and it is open to doubt if the mesonephros might not appear in later stages which were lacking among Price's materials.

Up to the oldest embryo observed by Price there were neither glomeruli nor bloodvessels of a definite form, although there were found in the splanchnopleure some vessels whose position seemed to suggest their corresponding to the glomeruli of Selachia and Amphioxus; "but they do not have any relation to the openings of the tubules, nor have they any direct connection

with the aorta" (loco cit., p. 213). The glomerulus figured in his Taf. 17, fig. 12 (yl) corresponds, I think, with a part of the glomerulus in Petromyzon.<sup>1)</sup>

As is very well known, the independent studies of Weiss ('90) and Boveri ('92) on the branchial chamber of Amphioxus gave a new direction to the morphological investigation of this field. A number of external openings of ciliated tubes is found at the dorsal corner of the peribranchial chamber of Amphioxus. Through the morphological study and physiological experiments this organ-system is demonstrated to be the excretory apparatus, or "Nierencanälchen," as Boveri calls them, of Amphioxus.

Boveri counted 91 "Nierencanälchen" in an individual 4 cm. in length and possessing 183 gill-bars on the right side. In the adult he counted about 180 of the "Nierencanälchen"; the number is, however, by no means constant, but varies within a certain limit.

In the middle region of the branchial chamber, a "Nierencanälchen" has 3 or 4 "Seitentrichter," and 2 "Endtrichter;" such is the most complete one. It becomes gradually simplified both anteriorly and posteriorly, until it is at last represented by a short single tubule, as seen in Taf. 33, figs. 9 and 13, given by BOVERI. The tubules in the anterior and posterior part of the system thus show a sign of degeneration, as in the case of the pronephros of Cyclostomata.

<sup>1)</sup> Dean has published two papers on the development of the Californian Hag ('98 and '99); these excellent works contain merely the general account of the course of the development in surface view. We may expect that the full account will throw much light on the ontogeny of Craniota. There stand, in the account given by him in these works, the important facts that the "pronephric tubules are apparent in connection with all the mesoblastic somites" ('98, p. 274) and that the pronephros is extended far backwards, beyond the anal region, into the tail ('99, p. 272). It would be highly desirable to observe the pronephric tubules of the Hag in relation to the myomere, and not to the spinal gaughia alone, as Price has done.

These nephric tubules receive, according to Boveri, the blood from the aorta, which gives two branchlets to each nephric segment. These branchlets form in each segment a network in the neighbourhood of, and winding around, the nephric tubule; it is this network that Boveri ealls glomerulus.

From the structure, the position, the segmental arrangement, the physiological function, and the relation of the blood-vascular system to this system of organs, Boveri regards the latter as a primitive form of Vertebrate nephric organ and homologised it particularly with the pronephros of Craniota. The points of difference which exist between the "Nierencanälchen" of Amphioxus and the pronephros of Craniota, have been smoothed away by the author's masterly arguments. The first of these points is the want of the segmental duct in Amphioxus; but this is represented, according to Boveri, by a part of the peribranchial chamber. The second is the relation of the nephric segments to other systems of organs. The "Nierencanälchen" is branchiomeric while the pronephros of Craniota is invomeric, in arrange-But this difference is looked upon by him as only apparent; for the number of gill-slits first formed agrees with that of the muscle-segments in the same region; this is sufficiently demonstrated by the figure given by Weiss (loco cit., fig. 3).

Thus the author has brought the "Nierencanälchen" of Amphioxus into perfect harmony with the pronephros of Craniota. Some additional light is now, I believe, thrown from the side of Craniota by the facts obtained in Cyclostomata, the lowest class of Craniota. This harmony will be brought out more in discussing the development of the pronephros in Selachia, Teleostei, and Amphibia, which will be treated further on.

388 s. hatta:

Thanks to the labors of many eminent investigators, the early development of the Selachian pronephros has been, as is well known, fully studied, so that the facts gathered from this field are well adapted to be compared with those from other groups. I have found, in the present investigation, many important points running parallel with the development of the Selachian pronephros. I may then be allowed to compare my own results in *Petromyzon* with those already arrived at in Selachia. Reference will, however, be limited to those works which are sufficient to verify the points 1 wish to bring out.

Through the excellent work of Ruckert ('88) we can best learn the origin of the pronephros in Selachia. "Die erste Anlage der Vorniere" is recognised "in Form einer gegen den Ectoblast gerichteten Vorbuchtung des parietalen Mesoblasts." This Anlage is first brought about by the thickening of the parietal layer of the mesoblast, which is found "in den Bereich des segmentirten Mesoblasts, d.h. Somiten " (p. 209); this thickening is called by the author "Segmentalwulst." The foot-note also runs as follows: "Der Ursprung des Segmentalwulstes reicht ventral bis zu der Stelle herab, wo die Somiten in den unsegmentirten Mesoblast der Peritonealwand übergehen" (p. 209). The "Segmentalwulst" is so called because it is noticed as the segmental thickening of the parietal mesoblast of which Ruckert recognised, in his Stad. 1111, six for Torpedo and four for Pristiurus, stretching over a corresponding number of the myotomes. The first indication of the pronephros is expressed, in Selachia also, segmentally in the segmental part of the mesoblast at the stage in which the metameric segmentation of the mesoblast is still going on, and

<sup>1)</sup> The embryos in the stage have 25-27 somites.

the myotome is not yet cut off from the lateral plate, just as in *Pctromyzon*. The foremost of them is found in the hind part of the third or fourth body-somite. The development of the Anlage in each segment agrees also with that in *Pctromyzon*; for he says: "Der Segmentalwulst zeigt in vorliegende Stadium (Stad. 1) regelmässig die stärkste Entwicklung in seiner mittleren Abschnitt, also ungefähr im Bereich des dritten des ihm gehörigen Somiten, und verjüngt von da allmählich nach seinem vorderen und hinteren Ende zu......" (loco cit., pp. 210).

The account given by Rückert is essentially confirmed by later investigators such as VAN WYHE ('89)11 RABL ('96), and others, although they differ from one another in the interpretation of the facts and in some unimportant points. RABL looks upon the Anlage of the pronephros (his Vornierenwulst) as the ventral portion of the somite just as Rückert does, while it is, according to VAN WYHE, the product of the lateral plate (his Hypomer). This is, as it seems to me, not a contradiction in the facts, but in the terms used; for VAN WYHE states: "Da nun der Pronephros, wie spätere Entwicklungsstadien zeigen, ein Produkt der Seitenplatte ist, während der unmittelbar dorsal davon liegende Theil des Mesoderms zur Mittelplatte gehört, ist die Segmentirung des Mesoderms bei Selachiern also nicht auf die Myotomplatte beschränkt, sondern erstreckt sich auch auf die Mittelplatte und den dorsalen Theil der Seitenplatte " (loco cit., pp. 474-475). The fact is, therefore, no other than that the portion of the mesoblast dorsal to the ventral limit of the Anlage of the pronephros undergoes segmentation, and the portion ventral to this point remains unsegmented, constituting the lateral plate. I will, in

<sup>1)</sup> The embryo, in which the first traces of the pronephros is seen, is, according to VAN WYHE, in a stage with 27 somites, whereas RABL has seen in an embryo of Pristiurus with 25 somites.

this place, not go further, but return in future pages to the discussion of this point. It is, however, safe, I believe, to regard this portion of the mesoblast as a part of the somite.

Van Wyhe found the foremost pronephric segment in the third body-somite (his Rumpfsegment), and Rabl states that the Vornierenwulst begins in the seventh somite formed (his Gesammtsegment). According to Rabl, however, van Wyhe's third Rumpfsegment corresponds to his seventh Gesammtsegment. To verify this fact Rabl has extended the comparison over Petromyzon, and found that in this case also the pronephros begins in the seventh somite; but the pronephric tubule in that somite is, as noticed above (p. 380), not the anteriormost of the tubules in his sense, but of the persistent tubules.

Van Wyhe noticed five of the pronephric segments for Raja, and three for Scyllium and Pristiurus; while Rable counted eight Vornierenwülste for Raja, and four for Pristiurus. The results in Petromyzon, therefore, best agree with those made out by Rückerr in Torpedo.

The authors agree in deriving the collecting duct from the lateral extremities of the pronephric Anlagen, where they become confluent.

RÜCKERT has observed, in *Pristiurus*, as well as in *Torpedo*, the *secondary connection* of the Segmentalwulst with the epiblast, which has led him to believe in some contribution of epiblastic cells to the formation of the pronephros, while VAN WYHE and RABL deny this. I have found the same connection in *Petromyzon*, but I have found no sign of the contribution of epiblastic cells to the formation of the pronephros. The phenomenon is temporary in both Selachia and *Petromyzon*; it takes place in Selachia, according to RÜCKERT, in his Stad. II,

and already in his Stad. III, a space is seen between these two structures.

The degeneration of the tubules in Selachia runs a course parallel with that mentioned under *Amphiocus* and Cyclostomata. As seen above, the Anlagen of the pronephros are developed most vigorously in the middle part of the pronephros, as in the case of *Amphioxus* and Cyclostomata; and degeneration begins at the cranial and caudal extremities as there.

Van Wyhe says that the degeneration consists in a confluence (Verschmelzung) of the ostia. According to Ruckert, the Vornierenfalte becomes simply flattened out in the cranial part of the pronephros. The reduction in the caudal part is noteworthy: the Anlagen are here constricted off from the mesoblast and converted into the anteriormost section of the segmental duct. Only the middle (the third) diverticulum (in *Torpedo*) persists in communicating with the body-cavity and becomes the *ostium abdominale*.

In Petromyzon, I have unfortunately failed to observe accurately the manner of degeneration of the tubule in the cranial part. It is however probable that it begins either from the blind tip of the tubule (the first tubule), or by obliteration of the nephrostome (the second tubule). In the caudal part, the collecting duct is constricted off from the lateral plate by obliteration of the tubule and constitutes the foremost section of the segmental duct, in precisely the same manner as in Selachia. The difference is: in Petromyzon the communication with the body-cavity is retained by the three middle nephrostomes, while in Selachia, it is through only the middle one, that is, the ostium abdominale.

The segmental duct becomes apparent in an embryo with

392 s. hatta:

35 (VAN WYHE), or 34 to 35 (RABL) somites. The anterior small section of the duct is formed, as just stated, in the same manner in Petromyzon and Selachia. The mode of formation of its posterior larger portion in Selachia differs from that of Petromyzon. RÜCKERT ('88) and VAN WYHE ('88, '89, '98) believe that it is the product of the epiblast1), while Rabl maintains its purely mesoblastic origin. At any rate, the posterior tip of the duct or the cord is sharply pointed and connected firmly with the epiblast throughout its growth until it opens into the cloacal cavity, which is effected, according to VAN WYHE and RABL, in the embryo with 83 to 84 somites. It can be inferred from VAN Wyhe's figs 7a and 7b, that this communication is found in a plane vertical to the thirty-eighth Rumpfsegment<sup>2</sup>. Petromyzon, the duct, being formed of a series of abortive pronephric tubules, has no genetic relation to the epiblast except in the cloacal region where the duct seems actually to receive cells from the epiblast, as fully stated above (p. 366).

The nephric arteries of Selachia which were discovered by Paul Mayer without reference to their relation to the pronephros, were studied by Ruckert and their true nature was pointed out by him. There are six of them in *Torpedo* corresponding to the number of the nephric segment; they are, however, not somitic but intersomitic in position. The vessels not only pass through the nephric fold, but throw a solid process, the interior of which consists of round or spindle-shaped cells. This is, according to Rückert, the equivalent of the pronephric glomerulus of Amphibia described by Fürbringer. The development and decline of these vessels go on parallel with those of the pronephric diver-

<sup>1)</sup> I will return to this point again in future pages.

<sup>2)</sup> According to Rabl's counting, this somite corresponds to his forty-second somite.

The vessels in the cranial as well as in the caudal part tienla of the pronephros are weaker than those in the middle (the third and fourth); only the latter vessels develop further and become the vitelline artery. Van Wyhe confirms Rückert's account and has described three vessels in Pristiurus. In addition to these, VAN WYHE has pointed out the very small segmental vessels on the left side, which go not to the intestine, but to the bodywall. They are not equivalent to the intestinal vessels on the opposite side. One of them gives a branchlet to the glomerulus which sends out, in its turn, a branchlet to the cardinal vein. The homologous vessels on the right side are to be seen coming out of the root of the vitelline artery. Bovers remarks that the vessels of Paul Mayer present many points of harmony with the branchial vessels in Amphioxus. Rabl agrees essentially with the account given by Rückert and VAN WYHE, but denies the existence of a glomerulus. According to RABL, the structure called the glomerulus by Rückert does not fulfil the conditions of being a glomerulus; he says: "Eine einfache Ausbuchtung einer Arterie ist noch keine Gefässschlinge, geschweige denn ein Glomerulus" (loco cit., p. 668).

Most of the early investigators, who observed the development of the Teleostian pronephros, believe it to be mesoblastic in origin. There are very few writers as Ryder ('87), and Brook ('88), who derive the segmental duct from the epiblast. According to Oellacher ('73), Goette ('75 and '88), Fürbringer ('78), and Hoffmann ('86), the first Anlage of the pronephros is brought about by the evagination of the parietal layer of the mesoblast at the level of the junction of the somite with the lateral plate, forming thus a longitudinal groove on each side,

which is subsequently constricted off from the body-cavity. This takes place at first in the middle region of the body, whence it proceeds both anteriorly and posteriorly.

Oellacher observed that the Anlage is converted into a longitudinal canal or the segmental duct, being completely shut off from the body-cavity in both the anterior and posterior parts. The anterior section of the duct is much swollen and transformed into the pronephric chamber. From the dorsal aorta, a pair of branches is given off which pushes into the pronephric chamber, pressing against its median wall and giving rise to a pair of the glomeruli. This portion of the duct becomes coiled up and constitutes the pronephros.

Goette's view somewhat differs from the account given above: the anterior end of the longitudinal groove is not completely closed from the body-cavity, but leaves awhile the communication with the latter, which is, according to Goette, the morphological equivalent of the nephrostomes of the Amphibian and *Petromyzon* pronephros. Opposite this nephrostome, he says, the glomerulus is formed by evagination of the visceral peritoneum and projects freely into the body-cavity. This portion of the peritoneum together with the nephrostome is constricted from the rest of the peritoneum; the coelomic cavity thus shut off is converted into the pronephric chamber.

This view is essentially confirmed by subsequent writers such as Furbringer ('78), Hoffmann ('86), and others, although Hoffmann differs in his view of the mode of the formation of the glomerulus.

According to the results recently arrived at by Felix in

<sup>1)</sup> I know his paper only by the abstract in the Jahresberichte über die Fortschritte der Anatomie und Physiologie, N.F. Ed. 111. '97.

the embryos of Salmonidæ, the earliest traces of the pronephros consist, in embryos with 11 pairs of the somites, of five solid proliferations of the lateral plate which is already cut off from the somite. These proliferations, being coincident with the caudal half of the third to seventh somites, are strictly metameric in arrangement and are regarded by the author as the rudimentary pronephric tubules. These tubules soon become confluent with one another to form a single outgrowth of the lateral plate, which is called by the author the "primare Vornierenfalte." The " primäre Vornierenfalte," which passes over into the parietal and visceral layers of the lateral plate, undergoes a longitudinal constriction (the "sekundäre Vornierenfalte") by which it is divided into the dorsal and ventral parts. From the former, the anterior section of the segmental duct originates, while the latter is transformed into the pronephric chamber. By stages, the dorsal part wanders laterally, and the ventral part travels medianwards. At the same time, these parts are separated from each other, leaving the communication at only one point, which is called the "Pseudonephrostom."

This phase of the development of the pronephros observed by Felix is, as I believe, undoubtedly earlier than that looked upon by the previous authors as the earliest indication of the pronephros.

At the time when the Anlagen of the pronephros are converted into the "Vornierenfalte," the Anlage of the caudal continuation of the segmental duct, becomes apparent in the eighth to the tenth somite; it is brought about by the division of the primary lateral plate (lateral plate in the ordinary sense) into (1) the secondary lateral plate (lateral), (2) the segmental duct (middle), and (3) the Anlage of the "Stammvenen" (median). This pro-

396 s. hatta :

cess proceeds posteriorly until the duct comes to lie close to the rectum (Enddarm).

Felix thus observed the segmental Anlage of the pronephros in its glandular part, and derives the rest of the system from the proximal margin of both the parietal and visceral layers of the secondary lateral plate; he has observed neither the posterior growth, nor the epiblastic origin, of the segmental duct.

The pronephric chamber which results from the confluence of the five pronephric tubules, is not homologous, according to Felix, with that of Amphibia, in which the chamber should be a constricted part of the body-cavity into which the tubules open.

Quite recently, Swaen and Brachet ('99) have published a paper on the early development of the mesoblastic organs in Salamonidæ. Although my manuscripts were nearly finished, when I saw this interesting paper, I must here refer in a few words to it<sup>1)</sup>.

The authors found the first traces of the pronephros under the fifth somite, of two embryos, one of which was in the stage of 11 somites, and the other of 13 somites. It is not the product of the parietal layer of the lateral plate only, but is formed, as Felix believes, by the proximal portion of both the parietal and visceral layers of the secondary lateral plate (l'extremé interne de la plaque latérale secondaire<sup>29</sup>). The internal cavity enclosed by the pronephros is, therefore, not the diverticulum, but a part of the body-cavity.

<sup>1)</sup> I am much indebted to my friend, Dr. A. Oka, who read the paper for me.

<sup>2)</sup> According to the authors, the "plaque latérale primitive" is divided into the "plaque latérale secondaire" and the "masse intermédiaire;" therefore, the "plaque latérale secondaire" corresponds to the lateral plate itself of *Petromyton*.

The Anlage of the pronephros is laid in exactly the same manner from the fourth somite to the cloacal region. Under the anterior three somites from the fourth to the sixth, the Anlagen are developed into the pronephric chamber; the Anlagen posterior to these are all transformed into the "canal excréteur," as they call the segmental duet, and they have come to the conclusion that the "canal excréteur" of the pronephros has the morphological value of a rudimentary pronephric chamber.

The facts given in the last two papers, are thus in close accordance with one another as well as with those given by myself in the foregoing pages. Differences between their results and mine are that the authors derive the system from the lateral unsegmented mesoblast, and that both the parietal and visceral layers of it partake in the formation of the system. As has been stated in the descriptive part, this derivation is only apparent; a little further study shows that only the parietal layer gives rise to the system, and this part of the layer belongs to the somite. Indeed, this part appears to form, for some time, the proximal portion of the lateral plate, being early cut off from the rest of the somite. It must be remembered that this separation is not the separation of the lateral plate from the somite, but that of the Anlage of the pronephros from the rest of the somite; or, the result of the development of the pronephros. is merely for a physiological reason that this development or separation of the pronephric Anlage goes on earlier than, for instance, in Selachia, it performing in Teleostei the actual excretory function. This will be understood easily, when a comparison with other groups is made further on.

It has been a well known fact that the development of

398 s. hatta :

Amphibia shows, in several respects, a parallel course with that of *Petromyzon*. Careful observations on the development of Amphibian pronephros, adduced by recent investigators, have intensified this similarity with the exception of a few points which are, however, probably of secondary importance.

Most authors who have worked on the Amphibian development agree in deriving the entire system of the pronephros from the parietal layer of the mesoblast only, and in regarding it as arising originally as a common pouch, the anterior part of which is divided secondarily, by a partial closure of the peritoneal communication, into a number of the pronephric tubules.

This view has been advanced by earlier authors such as W. Müller ('75), Goette ('75), Fürbringer ('78), Hoffmann ('86), and others. The stage at which the pronephros appears coincides exactly with that in *Petromyzon*, as Max Fürbringer says in his well known work: "Die erste Entwicklung der Vorniere und ihres Ausführungsganges findet hier nach der Scheidung des Mesoderms in Urwirbel und Seitenplatten statt und folgt unmittelbar der beginnenden Sonderung der ersten in einzelne Urwirbel amd der Spaltung der letzteren in Haut- und Darmfaselplatten. Embryonen von *Rana temporaria* von *circa* 2.5 Mm. Länge und von *Triton alpestris* von ca. 2.0 Mm. L. entsprechen diesen Stadium" (p. 3)<sup>11</sup>

MOLLIER ('90) has made out the segmental Anlage of the Amphibian pronephros, having worked with the embryos of *Triton*,

<sup>1)</sup> The nephrestomes are found, according to the author:

<sup>2</sup> in Salamandrina maculata,

<sup>2</sup> in Triton alpestris,

<sup>2</sup> in Siredon pisciformis.

<sup>3</sup> in Rana temporaria,

<sup>3</sup> in Bombinater igners (GOETTE), and

<sup>4</sup> in Coecilia rostrata (Spengel).

Bufo, and Rana. His accounts confirm, as a whole, those given by Rückert for Selachia above referred to, but differ somewhat from those of most other authors who have worked on Amphibian pronephros. Mollier states as follows: "Wir sehen hier ebenfalls zuerst eine solide, von dem Mesoblast ausgehende Anlage, deren Structur anfänglich schwer zu erkennen ist und erst mit dem Hohlwerden, wie bei den Selachiern, klar hervortritt. Dann finden wir, dass hier zwei resp. drei getrennte Canälchen vorhanden sind, die von den Somiten in convergenter Richtung ausgehen und erst nachträglich untereinander vereinigen zu einem Längscanal, von dem aus die Vornierentrichter in die Leibeshöhle führen" (loco cit., p. 229). The author derives in this wise the pronephric tubules, exactly as in the case of Petromyzon, from the segmented part of the mesoblast only.

Mollier's accounts are for the most part in close accord with the results given by Field ('91, p. 282), who, one year later independently of Mollier, began with Anura, and extended the work over Urodele Amphibia. In one point, their results differ widely; but "the difference is," it seems to Field, "apparent rather than real." According to Mollier, the nephrostomes communicate with the cavity of the myotome, the myocælome of van Wyhe; this is denied by Field, who believes that "the pronephric tubules have to do with the ventral segment of the mesoderm" (loco cit., p. 283). It seems to me that this "ventral segment of the mesoderm" corresponds to the pronephrotome of van Wyhe in Selachia or to "Pextremé interne de la plaque latérale secondaire" of Swaen and Brachet in

<sup>1)</sup> It seems that the earliest traces of the pronephros are perceived in an embryo younger than that with 7 somites.

Teleostei, and I agree with the view of Ruckert, here represented by that of Mollier.

Mollier and Field agree with each other in assigning three pairs of the tubules for Rana and Bufo, extending from the second to the fourth somite, and two pairs for Triton (Mollier) and Amblystoma (Field), covering the third and fourth somites. 
In addition to these, Mollier observed occasional occurrences of the third tubule in Triton, which is, according to Field, not equivalent, as Mollier maintains, to the third tubule in Bufo and Rana, because the additional third tubule in Triton is found in the fifth somite, while the third tubule in Rana and Bufo is under the fourth somite. According to Semon, there are ten pairs of the tubules on either side of the body in Ichthyophis.

A pair of glomeruli has been made out in Amphibia; the structure is connected by special vessels with the dorsal aorta on one hand and with the cardinal vein on the other. This branch of the aorta is believed by Field to correspond to a part of Mayer's vessels in Selachia. Beside these, there is no vessel arranged segmentally or otherwise.

The section of the body-cavity corresponding to the pronephric stretch is gradually expanded, and is shut off temporarily from the rest of the cavity by a close contact of the parietal and visceral layers of the coelome; this part of the cavity is, according to Goette, homologous with the pronephric chamber in Teleostei and with the homologous structure in *Petromyzon*, which is called by him the "peritoneale Scheidewände."

The so-called ventral portion of the Amphibian pronephros is, according to Mollier, brought about by the separation of

<sup>1)</sup> According to Field, Mollier's first body-segment in *Triton* corresponds to his third somite in *Amblystoma*.

the ventral portion of the pronephric Anlagen from the dorsal, which latter is differentiated into the tubules and constitutes the dorsal portion of the pronephros. The ventral part of the Anlagen separated from the dorsal retains anteriorly its connection with the anteriormost tubule and posteriorly with the segmental duct. It is prolonged and bent out anteriorly in front of the dorsal part. "Mollier's description is," Field says, "substantially in accord with my own observation,".........(loco cit., p. 286). This feature of the duct shows, it seems to me, a close resemblance to the anteriormost section of the Teleostean segmental duct which is, as above referred to, bent in the same fashion.

The segmental duct arises, according to previous writers, as a longitudinal common furrow oft he parietal peritoneum, which furrow is later constricted off from the mother-layer and becomes converted into a long canal. Mollier has observed the segmental duct transformed directly from the mesoblast, just like the glandular part of the pronephros, in the two somites behind the pronephros. Whether the greater remaining part of the duct is formed likewise by differentiation of the mesoblast, or by a backward growth of the hind end of the duct first formed, he could not decide with certainty; but the observations of Field elucidate this point.

"The segmental duct arises," FIELD says, "throughout its entire length by a proliferation in situ of the somatopleure" (loco cit., p. 223). The author has observed neither its epiblastic origin nor a free growth of its posterior end, except in the cloacal region where it "grows across the cloaca free from adjacent tissue" (loco cit., p. 223). In Stage v, the cloacal opening is seen. This opening is found, in Rana and Bufo, in the vertical plane with the middle of the twelfth somite, whereas it is below the twentieth somite in Amblystoma (FIELD).

402 s. Hatta:

The duct is segmental in origin. FIELD says: "I believe I am justified in concluding that the segmental duct between Somites v, and ix, arises in situ from a thickening of the somatopleure serially equivalent to that from which in the anterior region the pronephros is developed" (loco cit., p. 219). There are no other Vertebrata which agree more with Petromyzon with reference to the development of the segmental duct, than Amphibia. Indeed, here as there, the segmental duct is of segmental origin and is to be looked upon, as seen in Petromyzon, as the continuation of a series of abortive pronephric tubules in the posterior region.

Authors who have observed the epiblastic origin of the segmental duct in Amphibia are very few. von Perenji ('87) has published the results of his study on Rana esculenta, but his note is unfortunately very short. This view is opposed, so far I am aware, by almost all recent observers. After bringing the results by him into harmony with those by Rückert in Selachia, Mollier says: "Im einen Punkte weichen die Amphibia von Selachiern ab, dass die Vorniere mit dem Ektoblast in keine nähere Beziehung tritt. Allerdings heftet sie besonders in den Stadien, in welchen sie voluminöser erscheint, dem Ektoblast oft in affällend inniger Weise an. \* Doch lässt sich stets eine scharfe Grenze beiderlei Blätter ziehen, wenigstens bei Bufo, wo die Ektoblastelemente durch ihren Pigmentgehalt deutlich gekennzeichnet sind" (loco cit., p. 229).

The historical review undertaken in the foregoing pages shows the agreement to a large extent of the results arrived at in several groups of Anamnia. Some points of disagreement are naturally met with; but these are, I believe, only apparent.

<sup>1)</sup> I have not seen the paper by Brook.

In the groups above referred to, the first indication of the excretory system becomes apparent at a stage in which some mesoblastic somites are formed and the metameric segmentation of the mesoblast is going on. This is the Anlage not of the segmental duct, but of the pronephros. A single exception is found in Bdellostoma, in which the early traces of the system become visible, as we learn from Price, at a stage much more advanced than in other Anamnia, that is, at the stage in which the seleromyotome is cut off from the rest of the mesoblast and mesenchymatous cells fill up the spaces between organs and organ-systems.

I have endeavoured to reconcile the points, in which the views of the previous authors diverge from one another, under the following three headings:—

## 1.—The Anlage of the Pronephric Tubule is the Product of the Mesoblastic Somite and not of the Lateral Plate.

The view that derives the pronephros from a single common groove formed either of only the parietal, or of both the parietal and visceral layers of the unsegmented mesoblast (the lateral plate), is advocated by most of the authors who have worked on the development of *Petromyzon*, Teleostei, and Amphibia. This is due probably to the early separation of the sclero-myotome from the rest of the mesoblast in these groups. In them the Anlagen of the pronephros (or the nephric segments) together with the lateral plate are cut off from the sclero-myotome and form, for some time after this separation, the proximal portion of the lateral plate. It must be borne in mind that this separation is not the separation of the lateral plate, but of the nephrotome, from the sclero-myotome. This is, therefore, a

404 s. hatta:

step in the differentiation of the mesoblastic somite, and because the distal (ventral) portion of the latter happens for a time to be continuous with the lateral plate, we are not justified in concluding that it is derived from the lateral plate, which, as we know, never undergoes segmentation.

It is a significant fact that in Selachia and Amniota, in which the pronephros does not function as the actual excretory organ, this separation of the mesoblast into the selero-myotome and the nephrotome is not effected so early as in the above groups, but takes place only at later stages, with the first differentiation of the mesonephros. This consideration makes it reasonable to conclude that the early separation of the mesoblastic somite into the proximal and distal portions is caused by physiological necessity and has no morphological significance<sup>1)</sup>.

The case of Lacerta agilis is very instructive. According to Hoffmann ('89), the Anlagen of the pronephros in this animal are, in the most anterior segment, cut off from the myotome (selero-myotome) and remain connected with the lateral plate just as in Petromyzon, Teleostei, and Amphibia; whilst in all the following portion, they are the actual diverticula formed segmentally in the parietal layer of the lower part of the somite, as in other Reptilia (pp. 264 and 265). We thus see the two modes of separation in one and the same animal.

All recent authors agree in thinking that the Anlage of the pronephros is expressed in itself segmentally and is strictly myomeric. Now the question arises: How many parts are to be distinguished in the mesoblast, and to what part of it does the Anlage of the pronephros belong?

VAN WYHE ('89) has distinguished, in Selachia, three por-

<sup>1)</sup> This view is grounded upon the suggestion of Prof. Mitsukuri.

tions of the mesoblast which are called by him the "Epimer," "Mesomer," and "Hypomer" respectively. The epimere of VAN WYHE corresponds solely to the myotome; his mesomere comprises the Anlage of the mesonephros and the sclerotome; and the hypomere consists of the Anlage of the pronephros, the genital gland, and the lateral plate. The epimere, mesomere, and the dorsal part of the hypomere undergo the metameric segmentation, while the remaining portion of the hypomere remains unsegmented. According to VAN WYHE, the dorsal segmented part of the hypomere is, therefore, the product of the lateral plate (see p. 389). It seems to me that this division of the mesoblast does not agree with the facts observed in Petromyzon and the other Anamnia above referred to; for the mesoblast in these groups consists, in early stages, of two portions: (1) the segmented, and (2) the unsegmented, and of nothing more, just as RÜCKERT ('88) and RABL ('88, '96) have remarked. According to RÜCKERT and RABL, the segmented portion—the somite—comprises the myotome and the sclerotome; the pronephros and the mesonephros are derived from its ventral (distal) portion, which is called by Ruckert the "Nephrotom" ('88, p. 272).

In Petromyzon, these two portions of the mesoblast, the segmented and the unsegmented, are, in early stages, clearly distinguished, being histologically different (see p. 315). The mesoblast in such an undifferentiated state is almost entirely occupied by the segmented portion, while the unsegmented portion is very small, being represented by the loose tissue of a few cells. Such a mesoblastic segment exactly corresponds to the somite of RÜCKERT and RABL. The proximal half of the segmented portion coincides with the sclero-myotome of those authors. It consists not only of the myotome, but also includes the sclerotome. And it is the

406 s. hatta:

distal half of this segmented portion which folds out in each segment to give rise to the Anlage of the pronephric tubule on one hand and to the cœlomic projection on the other, and, therefore, corresponds to the "Nephrotom" of RÜCKERT.

The nephrotome, therefore, constitutes, in both *Petromyzon* and Selachia, precisely the same part of the mesoblast, viz. the distal (ventral) portion of the somite, through which the scleromyotome is connected with the lateral plate.

The above early stage in the differentiation of the mesoblast, in Petromyzon corresponds also to the "Ursegment" of Amphioxus in Hatschek's sense ('88). By further development of it the unsegmented mesoblast is brought into light, and we can then distinguish the "Urwirbel" and the "Seitenplatte" of Hatschek ('88). And the ventral half of the Urwirbel constitutest in Petromyzon, the connecting canal between the unsegmented ecclomic cavity and the sclero-myotome, that is to say, the nephrotome. Let us now examine what part of the Ursegment of Amphioxus represents the nephrotome of the Craniota.

In his excellent work on "Die Nierencanälchen des Amphioxus," Bovert ventures to solve this important question. After a discussion he comes to the conclusions:

- (1) That the "Gononephrotom" in Craniota must correspond to a part of the "Urwirbel" of Hatschek;
- (2) That the "Gononephrotom" of Craniota is homologous with the genital chambers of the adult Amphioxus.

But these chambers "sind urspränglich die segmentale Verbindungscanäle zwischen der unsegmentirten Leibeshöhle und der Sclero-Myotom gwesen" ('92, p. 493). I can, therefore, aseribe no other significance to the ventral half of the segmented mesoblast in *Petromyzon* than that it is the morphological equivalent of the "segmentale Verbindungscanäle."

It thus follows that the distal half of the segmented mesoblast in *Petromyzon* undergoes exactly the same fate as that in *Amphiorus*: it is transformed into the pronephros and the cœlomic projection or the "dorsal segmental cœlome," which latter gives rise, just as Boyeri suggests in *Amphiorus*, to the mesonephric tubules and, in the hinder region, to the genital gland.

As has been pointed out in the historical review, the relation of the Anlage of the pronephric tubule to the mesoblastic somite is the same for Teleostei and Amphibia, as in *Petromyzon*.

It may, therefore, safely be stated, that the segmented portion of the mesoblast constitutes in these groups a single integral structure until the separation of the nephrotome in continuo with the lateral plate from the selecto-myotome. This separation is, as above stated, not the separation of the somite from the lateral plate, but the differentiation of the somite into the selecto-myotome and the nephrotome, preparatory to the development of the urogenital system. The reason why the separation takes place earlier in some groups than in others, rests only on physiological grounds.

B.—The Whole System of the Pronephros of Cyclostomata,
Teleostei, and Amphibia is Homologous with the
Nierencanälchen of Amphiorus (Boveri) and
not perfectly Homologous with the Selachian
Pronephric System.

I have already stated above (pp. 386 and 387) that Boveri has brought the pronephric system of Craniota in harmony with the system of the "Nierencanälchen" of *Amphiorus*, basing his arguments on the structure, the position, the myomeric arrange-

ment, the physiological function, and the relation of the vascular system to the organ. His comparison is, however, almost entirely limited to Selachia on the side of Craniota, owing perhaps to the scantiness of the literature at that time. I accept in the main this homology, and I may perhaps extend this comparison a little further.

I will begin with the homology of the pronephros of Cyclostomata with the "Nierencanälchen" of Amphioxus.

It is well known that the starting point of the hepatic diverticulum from the enteric canal demarcates, in the Chordata, the respiratory section of the canal from the nutritious section of it; and, as Gegenbaur ('78, pp. 563—581), Balfour<sup>1)</sup> ('85), and others affirm, the esophagus and stomach in the higher forms are a part of the former section, which is called the fore-gut. And the homology of the hepatic energy of Amphiorus with the liver of the Craniota, has been much strengthened by recent morphological studies and physiological experiments<sup>2)</sup>. The results of my present study also confirm this view. I will use, therefore, this fixed point as the landmark of comparison of the two organ-systems, the pronephros and the "Nierencanälchen," and of the pronephros in different groups of Craniota.

<sup>1)</sup> From the account of Balfour, I will cite the following lines:-

<sup>&#</sup>x27;In Amphioxus the respiratory region extends close up to the opening of the hepatic diverticulum, and therefore to a position corresponding with the commencement of the intestine in higher types. In the craniate Vertebrata the number of the visceral cletts has become reduced, but from the extension of the visceral clefts in Amphioxus, combined with the fact that in the higher Vertebrata the vagus nerve, which is essentially the nerve of the branchial ponches, supplies, in addition the walls of the osophagus and stomach, it may reasonably be concluded, as has been pointed out by Gegenbaur, that the true respiratory region primitively included the region which in the higher types forms the oesophagus and stomach' (Vol. II, p. 758).

Balfour has also shown that the solid cord of the esophagus in Elasmobranchii and Teleostei, is the remanent of the gill-radiments in the ancestry (loco cit., pp. 61 and 78).

<sup>2)</sup> J. A. HAMMAR, '99, '98, and Guido Schneider, '99.

The "Nierencanälchen" of Amphioxus, according to Boyeri ('92), extends over and is limited to, the whole extent of the branchial region, the posterior larger part of which covers the hepatic cœcum. The pronephros of Cyclostomata extends from the anterior body-somite to the cloaca. The anterior section of the system constitutes afterwards the glandular part represented by the pronephric tubules and is found in front of, and over, the Anlage of the liver, or in the region of the fore-gut; a certain number (two in Petromyzon, twenty in Bdellostoma) of the anterior nephric segments are found in the branchial region, and the posterior one or two segments of the glandular part (Petromyzon) cover the liver-Anlage. It follows that the six10 to twenty or more pronephric tubules correspond to as many "Nierencanälchen" in about the middle one third of the branchial region of Amphioxus, and that the "Nierencanälchen" lying posterior to this point are represented, in Cyclostomata, by a number of the rudimentary tubules which are converted into the segmental duct.

The "Nierencanälchen" are not put in communication with one another by the collecting duct, as in the pronephros of Cyclostomata, but open to the exterior segmentally. I have stated in the descriptive part (p. 333) that the free extremities of the pronephric tubules in *Petromyzon* are brought into close contact with the epiblast, so that the latter is pressed out by the enormous growth of the tubule and that this is especially the case in the first and second tubules. This fact throws light upon the homology of the pronephric tubules in *Petromyzon* with the "Nierencanälchen" of *Amphioxus*: in other words, the condition

<sup>1)</sup> The glandular part of the pronephres in *Tetromyzon*, are represented by the six pronephric tubules.

<sup>2)</sup> The branchiomeres in the posterior section of the gill-basket of Amphioxus are afterwards added (see pp. 410-411).

seen in the "Nierencanälchen" would be brought about, if the tubules in *Petromyzon* came to open to the exterior, boring through the epiblast by the further growth of their free extremity. This intimate contact of the tubule-end with the epiblast takes place, as above mentioned, in the middle of Stage II, where the tubules have developed a little beyond the mere Anlage.

The pronephric tubules of *Petromyzon* are, in this stage, already united with one another by the intersomitic solid cord; this union is, however, not primary, but secondary. This stage presents, I think, the phylogenetic stage, in which the "Nierencanälchen" with separate external segmental openings, and the pronephric tubules with the collecting duct, diverge from each other.

By this assumption, it is not meant that in the ancestry of Chordata the tubules were closed blindly inside the epiblast; for the Anlage of the pronephric tubule might have been, in the ancestral form too, brought about by the folding of the mesoblast, to break out finally to the exterior. This perforation would become unnecessary when the secondary union of the tubules had been acquired.

Since a certain number of the "Nierencanälchen" in front of the base of the hepatic cocum, is represented by the pronephric tubules of the glandular part in Cyclostomata, those lying over it will be homologous with the pronephric tubules which are found over and posterior to the hepato-pancreatic Anlage and converted into the anterior section of the segmental duct, being secondarily united with one another by the confluence of the free extremities of the tubules.

There is not to be seen the post-hepatic "Niereneanälchen" in Amphiorus. We learn from Lankester ('89) and Willey ('91) that in Amphiorus, the new branchial slits are added, by stages,

to the posterior end of the pharynx, so that, in later stages, the coincidence of the number of the slits with that of the myotomes is lost; and that this addition of the slits continues throughout life. The nephrotomes in these new slits have been, I think, originally coincident, in each segment, with the myotomes, from which they were cut off in early stages and have remained undeveloped until the new appearance of the added slits. It seems, therefore, probable that the branchial region of Amphiorus once extended over the largest portion of the enteric canal, while a very small section in the posterior part of the canal performed the nutritious function, as is seen now in the Ascidian<sup>10</sup>. The "Niereneanälchen" in this hinder part may represent the pronephric tubules in the post-hepatic section of the segmental duet of Cyclostomata<sup>21</sup>.

The pronephric system of *Petromyzon* comes to have the same relations with the epiblast as the "Nierencanälchen" of *Amphioxus* at three different points: the free ends of the two anterior pronephric tubules and the hind end of the segmental duct (probably the hindmost pronephric tubule). Whilst in the greatest section of the system the communication with the exterior has been lost, these three points might have preserved it to a considerably later phylogenetic stage: the two anterior tubules playing the same physiological part as the "Nierencanälchen" of *Amphiorus*, and the posterior being employed as the only excretory pore of the system secondarily established by the union of the tubules.

In main points (with exception of the presence of the tubules in the branchial region, of the contact or connection of

<sup>1)</sup> Balfour says: "In Ascidians the respiratory sack is homologeus with the respiratory tract of Amphioxus" (10:0 e.7., p. 758.)

<sup>2)</sup> See p. 108.

them with the epiblast, &c.), the pronephric system of Teleostei and Amphibia shows, as stated in the historical review, the same characters as that of Cyclostomata, so that the facts established in Cyclostomata have the same significance for the Teleostei and Amphibia.

Although such is the case in those Craniota and Amphioxus, the pronephros of Selachia is quite otherwise: the Anlagen of the pronephros are here formed in the mesoblastic somites posterior to the Anlage of the liver (see below), and only one or two segments of them are converted into the segmental duet (Rückert). These pronephric Anlagen in Selachia are, therefore, the morphological equivalent, not of the glandular portion of the pronephros, but of those which are converted into the segmental duet in the Craniota just mentioned.

C.—The Segmental Duct in Selachia is not the Morphological Equivalent of the Duct of the Same Name in Cyclostomata, Teleostei, and Amphibia.

Contradictory views are met with in the derivation of the segmental duct. The results arrived at in Cyclostomota, Teleostei, and Amphibia, well agree in making it of the mesoblastic origin; there are a few authors who believe in the epiblastic origin of the duct in these groups, but their papers are not more than mere notes. In Selachia, the circumstance is reversed; I am not aware of any recent author other than Rabl, who advocates the mesoblastic origin of the Selachian segmental duct. The facts given by Rabl are, however, not the same as those observed in the groups just referred to. In these, as stated above, the duct is

differentiated, so to speak, in situ from the mesoblast in its whole length, and as recent authors agree, is composed of a series of the abortive tubules formed in each nephrotome. This is not the ease in Selachia; here it is brought about, as Rabl states, by the posterior growth of the collecting duct which is formed by the confluence of the lateral extremities of the pronephric Anlagen. It is not easy to bring these two widely divergent modes of formation into harmony with each other.

A few morphological considerations, however, would, I believe, enable one to derive one type of the system from the other. I may be permitted to state here some of these considerations.

I will start with the question: Is the segmental duct of Selachia the morphological equivalent of that of *Petromyzon*, Teleostei, and Amphibia? I believe the question can be answered safely in the negative, if we consider (1) the position of the pronephros first formed, and (2) the origin of the duct.

In the first place, the pronephros in Selachia appears, as we learn from Rabl, in the mesoblastic somites lying posterior to the Anlage of the liver; thus the Anlage of the liver lies under the fourth and fifth somites, and that of the pancreas under the sixth, while the pronephros covers the seventh to tenth somites ('96, p. 667). On the contrary, in Petromyzon<sup>1)</sup> the pronephros originates in the mesoblastic somites anterior to the hepato-pancreatic Anlage, only the posterior one or two nephrotomes covering the liver. Such being the case, the pronephric segments in Selachia correspond to the same number of the abortive tubules in Petromyzon and the other Craniota above

<sup>1)</sup> As we learn from Goette ('75) and Oellacher ('73), the anterior section of the pronephric system in Amphibia and Teleostei, is also found in the mesoblast opposite to the posterior section of the fore-gut.

mentioned, which are converted into the anterior section of the segmental duct in these groups. It follows that the segmental duct in this part of *Petromyzon* and the two other groups, is not the morphological equivalent of the duct of the same name, but of the pronephros itself, in Selachia.

In the second place, let us consider the mode of growth of the segmental duct. Whichever view may be taken of its origin, whether epiblastic or mesoblastic, this duct in Selachia does not arise segmentally as in the other Craniota just referred to. It is a backward growth produced either by delamination from the epiblast, as Rückert and van Wyhe affirm, or by cell-multiplication within the structure of the mesoblastic collecting duct itself, as Rabl states. Hence it can not be homologous with the duct of the same name in *Petromyzon* and the two other groups, which is derived segmentally from the rudimentary pronephric tubules. The Selachian segmental duct is, in its whole length, represented, as I believe, by the posterior small section of the segmental duct in *Petromyzon* and Amphibia.

In Petromyzon, the hind end of the duct comes into an intimate connection with both the epiblast and the lateral diverticula of the cloaca, filling up the space between them, and fusing with both of them. We may suppose that the direct communication of the duct with the exterior, if such truly existed in the ancestral history, may have been at this point of the epiblast. This fused condition of the duct and the epiblast reminds us of the early stages of the Selachian duct at the stage when it has been produced only a little posteriorly from the pronephric region. I believe that if the duct is to be compared in Petromyzon and Selachia, a stage such as the above ought to be taken. The largest part of the Selachian duct is represented by a free hind-

ward growth formed after such a condition is passed, and its homologue can not be found anywhere in *Petromyzon*. I believe that the same can be stated of Amphibia which is, to judge from Field's account, very much like *Petromyzon* in this respect (see p. 401).

According to VAN WYHE and RABL, the duct in Selachia appears in the seventh to the tenth (in Rabl's sense) somites of embryos with 34-35 somites and, when it is later connected with the cloacal wall, the connection is found,—as can be inferred from fig. 7h of van Wyhe,—in the thirty-eighth (forty-second of RABL) Rumpfsegment (or further backwards) of *Pristiurus* embryos with 80 (VAN WYHE) to 87 (RABL) mesoblastic somites. There is found in Selachia, therefore, a number of the mesoblastic somites in the region back of the pronephros, which do not give rise either to the pronephric tubules or to the segmental duct, and the duct grows backwards, free from the mesoblast inside, during the period in which the somites increase from 34-35 to 80-87, and for the space reaching from the eleventh or twelfth to the fortysecond (in the sense of RABL) somite. Such a considerable prolongation of the duct during this period is not observed in Petromyzon<sup>11</sup> and in the two other groups of Craniota mentioned.

And furthermore, it is questionable whether, during this posterior growth, the duct in Selachia receives the constituent cells from the epiblast along its whole length, as RÜCKERT and VAN WYHE believe; or only at the point of the epiblast overlying the hind end of the pronephros, with which the duct is connected, and posteriorly to this point grows free from both the epiblast

<sup>1)</sup> At about this stage (Stage IV), there is no space left behind the pronephric system segmentally formed; for the embryo of *Petromyzon*, is retort-shaped and has the anus situated in the ventral median line of the bulb of the retort.

and the mesoblast. The latter view seems probable to me. The figures ('89, figs. 5 a-c) given by van Wyhe to illustrate his view of the epiblastic origin of the duct, are from the vertical plane of the eighth Rumpfsegment of a Scyllium embryo with 37 somites, which corresponds to the twelfth Gesammtsegment of Rabl. The figures ('96, figs. 9a, 9b, 10a, and 10b) given by Rabl to the negation of van Wyhe's view, are from the vertical plane of the twenty-second Gesammtsegment of an embryo with the 63 somites. These two cases are, I suppose, the two ends of the same duct in different stages; the anterior end, being the equivalent of the hind end of the segmental duct in Petromyzon, actually receives cells out of the epiblast, as the figures by van Wyhe show; the other end, which is seen in Rabl's figures, is the point of mere contact with the epiblast, along which it is shifting backwards.

If the above comparison be correct, the segmental duct in Schachia is, except the anterior very small section which is formed directly of the abortive tubules, not homologous with the duct of the same name in Petromyzon, but is a structure secondarily acquired.

From the above account, it may be safely concluded that in its primary phylogenetic stage, the pronephric system of the Craniota above referred to consisted of a number of segmentally arranged tubules, which were directly formed, in each mesoblastic segment, from the distal (ventral) portion of the mesoblastic somite, and opened independently to the exterior: that the lateral extremities of these tubules were afterwards secondarily united with one another, thus constructing the collecting and segmental duct, the hind end of which opened directly to the exterior; and that the acquisition of an opening of the duct into the cloaca

was the tertiary stage of changes in the system. Such a course of the phylogenetic development of the system is, however, no other than that advanced by RÜCKERT ('88, p. 265).

In the present paper, the historical comparison will be limited to the groups of Vertebrata stated above; the review of Amniota and some other theoretical considerations will be reserved to a future paper, in which I propose to deal with the further fate of the pronephros and the development of the mesonephros in *Petromyzon*.

Having compared the results arrived at in the present work, with those in different classes of Anamnia, I may be justified in drawing the following conclusions.

In *Petromyzon*, the first indications of the pronephros becomes apparent at a stage earlier than those hitherto regarded as the starting point, that is, at a stage in which the mesoblast in the anterior region has undergone the metameric segmentation but the lateral plate is not yet cut off from the somite.

The tissue giving rise to the pronephros is the parietal layer of a small section of the mesoblast, which forms the distal (ventral) half of the mesoblastic somite. This section of the mesoblast exactly corresponds to the "Nephrotom" of Rückert in Selachia.

The Anlage of the pronephros in all the groups of Vertebrata above referred to is produced by the evagination of the parietal layer of the nephrotome which theoretically ought to contain a part of the colonic cavity. In Cyclostomata, such a cavity is 418 s. hatta:

actually present<sup>1)</sup>: in other groups, the Anlagen are mere thickenings.

As the pronephric tubules are derived, in each segment, from the distal (ventral) half of each mesoblastic somite, the pronephros is, from the first, of a segmental arrangement, being strictly *myomeric*. In fact, the separation of the sclero-myotome from the lateral plate is effected on account of the differentiation of the Anlage of the pronephros or of the nephrotome.

In *Petromyzon*, the pronephric tubules which constitute the glandular part of the system and the anterior section of the segmental duct, are formed in the region of the fore-gut and some of them are detected in the region where the gill-pouches are afterwards formed; these latter disappear entirely before the gills come into view.

The segmental Anlagen of the pronephric tubules are secondarily connected by the duct formed out of two adjacent pronephric Anlagen and put in communication with one another.

The degeneration of the pronephric tubules takes place from both the cranial and caudal extremities of the system. In the cranial part, the tubules disappear without leaving any trace; while in the caudal, they are converted into the anterior section of the segmental duct. The remaining part of the system functions for some time as the excretory organ.

The pronephric Anlagen in the hinder region do not develop beyond a certain point, but are employed solely to give rise to the segmental duct just as in the somites having degenerated tubules.

From what has been said, it is, I renture to think, no rash conclusion to regard the pronephric tubules in Petromyzon as having once extended over the body-segments from the branchial region

<sup>1)</sup> According to SWAEN and BRACHET, the same fact is seen in Teleostei.

to the cloacal part, and as having been, in the anterior region, replaced by gills and, in the posterior, converted into the segmental duct.

In the two anterior segments which belong to the branchial region, the free ends of the tubules are brought into close contact with the epiblast but this germinal layer has, in this region, no share in the formation of the system. The hind extremity of the segmental duct, however, strikes against the epiblast and has every appearance of receiving some cells out of it. These facts allow us to infer that all the tubules once had each an independent external opening until they were secondarily united with one another by the intersomitic duct.

The visceral layer of the nephrotome becomes evaginated medianwards and forms a series of segmental pouches on either side of the subchorda; but this feature is temporary, and the structure is soon smoothed by their becoming confluent with one another. This series of pouches is, I believe, the remnant of the *primitive segmental cælome*, and gives rise to the gonads and the mesonephros.

If the accounts given above be correct, the primary mesoblast is, during early development, divided into two distinct portions: (a) the larger proximal portion which is segmented, and (b) a small distal portion which is unsegmented. The former is differentiated into the sclero-myotome and the nephrotome, and the latter forms simply the peritoneal linings.

The pronephric vessels acquire their definitive form in much later stages; when established, they are intersomitic in position. The posterior part is transformed into a pair of the glomeruli of the pronephros.

Petromyton has for a long time been looked upon as being

420 s. hatta:

peculiar and standing apart from other Vertebrata in the development of the pronephros. But the results brought out in the present work speak for a complete parallelism between this genus and the representatives of other classes of Vertebrata.

Biological Laboratory,

The College of Peers, Tokyo.

November, 1899.

# Postscript.

By the kindness of Prof. Watasé, I have been enabled to look through Dr. Wheeler's paper on "The Development of the Urogenital Organs of the Lamprey" which has just been published. I find a general agreement of his results with mine. The most important point is the discovery of the earliest traces of the pronephros as given in the foregoing pages. As to the formation of the segmental duct, his views are somewhat different from mine; this and some other points of divergence are, as I believe, due to gaps in his materials. Thus, his fig. 1, which represents section through an embryo in his Stage 1, corresponds to my fig. 1, while the next older stages (Stages 2 and 3) in his series, spoken of as representing Goette's fig. 9 where the heart is already formed, coincide with the oldest embryo of my Stage 1v. As seen in the foregoing description, most of the important processes in the

<sup>1)</sup> Zool. Jahrbücher, Abtheil, für Anat, and Ontog., Bd. XIII, 799.

development of the pronephros and the segmental duct take place in this interval of time, which Wheeler has unfortunately omitted to study. But in the main, his results confirm mine. This agreement arrived at independently naturally affords a good evidence of the correctness of the facts given.



#### Literature.

- ('79) Balfour:—A Monograph on the Development of Elasmobranch Fishes.
- ('87) Beard:—On the Origin of the Segmental Duct in Elasmobranchs:
  Anat. Anz., Bd. 11.
- ('92) Boveri:—Die Nierencanälchen des Amphioxus: Zool. Jahrb., Bd. v.
- ('88) Brook:—Note on the Epiblastic Origin of the Segmental Duct in Teleostean Fishes and in Birds: Proceedings of the Royal Society of Edinburgh, Vol. XIV.
- ('98) Dean:—On the Development of the Californian Hagfish (Bdellostoma stouti Lockington): Quart. Journ. Micr. Sc., N. S., Vol. XL.
- ('82-'90) Dohrn:—Studien zur Urgeschichte der Wirbelthirkerpers: Mittheil. a. d. Zool. Stat. z. Neapel.
- ('91) Field:—The Development of the Pronephros and Segmental Duct in Amphibia:—Bullt. Mus. Comp. Zool. Harv. Coll., Vol. xxi.
- ('86) Flemming:—Die ektoblastische Anlage des Urogenitalsystems bei Kaninchen: Arch. f. Anat. u. Physiol., Anat. Abtheil.
- ('97) Felix:—Die Price'sche Arbeit "Development of the Exerctory Organs of a Mixynoid (Bdellostoma stouti)" und ihre Bedeutung für die Lehre von der Entwickelung des Harnsystems: Anat. Anz., Vol. XIII.
- ('78) FÜRBRINGER:—Zur Vergleichenden Anatomie und Entwicklungsgeschichte der Excretonisorgane der Vertebraten: Morph. Jahrb., Bd. iv.
- ('78) Gegenbaur:—Grundriss der Vergleichenden Anatomie, 2. Auflage.
- ('75) Goette:—Die Entwicklungsgeschichte der Unke, Leipzig.
- ('88) ——: Ueber die Entwickelung von P. planeri : Zool. Anz., Jahrg. VI.
- ('97) Gregory:—Origin of the Pronephric Duct in Selachians: Zool. Bullet. Vol. 1.
- ('81) Hatschek:—Studien über Entwickelung des Amphioxus: Arb. a. d. Zool. Inst. d. Univ. Wien., Tom. iv.

- ('98) Hammar:—Zur Kenntniss der Leberentwickelung bei Amphioxus: Anat. Anz., Bd. xv.
- ('97) ——:—Ueber einige Hauptzüge der ersten embryonalen Leberentwickelung: Id., Bd. xiv.
- ('88) Hatschek: Ueber den Schichtenbau von Amphioxas: Id., Bd. и.
- ('91) Hatta:—On the Formation of the Germinal Layers in Petromyzon: Jour, Coll. Sc. Imp. Univ., Jap. Vol. v.
- ('97) ——:—Contributions to the Morphology of Cyclostomata, I. On the Formation of the Heart in Petromyzon: Id., Vol. x.
- ('90) Hertwig:—Lehrbuch der Entwicklungsgeschichte des Menschen und der Wirbelthiere, 3. Auflage.
- (86) Hoffmann:—Zur Entwicklungsgeschichte der Urogenitalorgane bei den Anamnien: Zeitschrift f. wiss. Zool., Bd. xliv.
- ('88) v. Kuffer:—Ueber die Entwickelung der Neumaugen: SB. d. k. bayer. Akad. Wis. zu München, Bd. I. (Ref. in Jahresb. Fortschr. Anat. Phys., '89).
- ('90) ———:—Die Entwickelung von P. planeri: Arch. f. mikrosk. Auat., Bd. xxv.
- ('89) Lankester:—Contributions to the Knowledge of Amphioxus lanceolatus Yarrell: Quart. Journ. Micr. Science, N. S., Vol. XXIX.
- ('90) Lankester & Willey:—The Development of the Atrial Chamber of Amphioxus: Id., xxxi.
- ('97) Maass:—Ueber die Entwicklungsstadien der Vorniere und Urniere bei Myxine: Zool. Jahrb., Bd. x.
- ('88) Martin:—Ueber die Anlage der Urniere beim Kaninchen: Arch. f. Anatomie and Entwicklungsgeschichte.
- ('87) Mayer. (P.):—Ueber die Entwickelung des Herzens und der grossen Gefässstämme bei Selachiern: Mittheil. Zool. Stat. Neapel, Bd. vи.
- ('88) Mitsukuri:—The Ectoblastic Origin of the Wolffian Duct in Cheolonia (Prelim. Notice): Zool. Anz. Jahrg. xi.
- ('90) Mollier:—Ueber die Entstehung des Urogenitalsystems bei Amphibien: Arch. Anat. Phys., Anat. Abth., Bd. 111.
- ('75) Muller (W.):—Das Urogenitalsystem des Amphioxus und Cyclostomen; Jen. Zeitschr. Naturw.. Bd. 1x.

- ('72 & '73) Oellacher:—Beiträge zur Entwicklungsgeschichte der Knochenfische: Zeitsch. f. wis. Zool., Bd. XXII u. Bd. XXIII.
- ('70) Owsjanniskow:—Die Entwicklungsgeschichte der Flussneunaugen: Melanges Biologie, Tom. vii.
- ('87) V. Perenji:—Die ectoblastische Anlage des Urogenitalsystems bei Rana esculenta und Lacerta viridis: Zool. Anz., Jahrg. X.
- ('97) Price:—Development of the Excretory Organs of a Myxinoid (Bdellostoma stonti Lockington): Zool. Jahrb. Bd. x.
- ('88) Rabl:—Ueber die Differenzirung des Mesoderms: Anat. Anz., Bd. 111.
- ('87) ——:—Theorie des Mesoderms; Morph. Jahrb., Bd. xv.
- ('96) ——:—Ueber die Entwickelung des Urogenitalsystems der Selachier (zweite Fortsetzung der "Theoric des Mesoderms"): Id., Bd. xxiv.
- ('88) RÜCKERT:—Ueber die Entstehung der Excretionsorgane bei Selachiern: Arch. Anat. u. Phys., Anat. Abth.
- ('89) —— :—Zur Entwickelung des Excretionssystems der Selachier; eine Erwiederung an Herrn van Wyhe: Zool. Anz., Jahrg. XII.
- ('82) Ryder:—A Contribution to the Embryography of Osseous Fishes with Special Reference to the Development of the Cod: Report U. S. Fishcommission.
- ('99) Schneider, (Guido):—Einiges über Resorption und Exerction bei Amphioxus lanceolatus: Anat. Auz., Bd. xvi.
- ('56) Schultze, (Max):—Die Entwicklungsgeschichte von Petromyzon planeri, Haarlem.
- ('82) Scott:—Beiträge zur Entwicklungsgeschichte der Petromyzonten. Morph. Jahrb., Bd. vii.
- ('81) Sedgwick:—On the Early Development of the Anterior Part of the Wolffian Duct and Body in the Chick: Quart. Journ. Micr. Sc., N.S., Vol. XXI.
- ('96) Semon:—Das Excretionssystem der Myxinoiden in seinen Bedentung für die morphologishe Auffassung (Ref. Jahresber ü. d. Fortsch. d. Anat. u. Phys., N. F., Bd. 11).
- ('97) ——:—Das Excretionssystem der Myxinoiden: Anat. Anz., Вd. vии.
- ('97) ——:—Vorniere und Urniere, Id., Bd. XIII.
- ('75) Semper:—Das Urogenitalsystem der Plagiostomen und seine Bedeutung für das der übrigen Wirbelthiere: Arbeiten a. d. zool.-zootom. Inst. in Würzburg, Bd. 11.
- ('87) Shipley:—On Some Points in the Development of Petromyzon fluviatilis: Quart. Journ. Micr. Sc., N.S., Vol. xxvu.

- ('95) Sobota :—Die Entwickelung der Vorniere der Salmoniden : Anat. Anz., Bd. vi.
- ('84) Spee, (Graf):—Ueber directe Betheiligung des Ektoderms an der Bildung der Urnierenanlage des Meerschweinchens: Arch. f. Anat. und Phys., Anat. Abth.
- ('99) SWAEN et BRACHET:—Entude sur les premières phases du dévelopment des organs dérivés du mésoblaste chez les poissons téléostéeus: Arch. de Biologie, Tom. xvi.
- ('90) Weiss:—Excretory Tubules in Amphioxus lanceolatus: Quart. Journ. Micr. Sc., N. S., Vol. xxx.
- ('84) Weldon:—On the Head-kidney of Bdellostoma, with a Suggestion as to the Origin of the Suprarenal Bodies: Quart. Journ. Micr. Sc., N.S., Vol. xxiv.
- ('83) Wiedersheim:—Lehrbuch der vergleichenden Anatomie der Wirbethiere.
- ('91) WILLEY:—The Later Larval Development of Amphioxus: Quart. Jour. Micr. Sc., N.S., Vol. xxxi.
- ('86) v. Wyhe:—Die Betheiligung des Ectoderms an der Entwickelung des Vernierenganges: Zool. Anz. Bd. 1x.
- ('89) ——:—Ueber der Mesodermsegmente des Rumpfes und die Entwickelung des Excretionssystems bei Selachiern: Arch. f. mikr. Anat., Bd. xxxIII.
- ('88) Ziegler:—Der Ursprung der mesenchymatischen Gewebe bei den Selachiern: Arch f. mikr. Anat., Bd. xxxii.





## Plate XVII.

[The magnification is the same for all figures:  $C \times 2$ , with the single exception of fig. 4 which is  $E \times 2$ .]

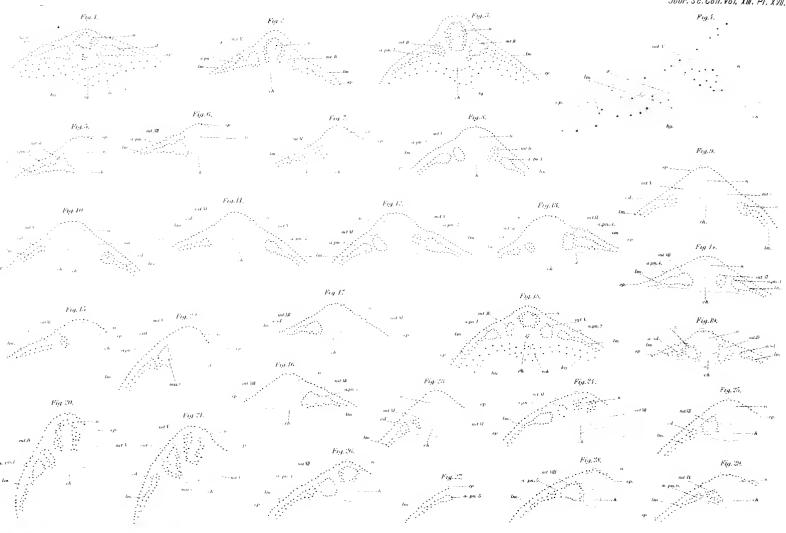
a.pn.1-6, Anlagen of pronephric tubules from the first to sixth.
a.sd., Anlage of segmental duct.
cd., collecting duct.
ch., chorda dorsalis.
cut., cutis-layer of myotome.
d., dorsal row of mesoblast.
cp., epiblast.
hy., hypoblast.
l.m., lateral plate of mesoblast.

m., median row of mesoblast. m.p., parietal layer of mesoblast. mes., mesoblast.

mt.I, II, &c., the first, second, &c. myotome.

mus., muscle-layer of myotome.
m.v., visceral layer of mesoblast.
n., neural cord or canal.
v., ventral row of mesoblast.

- Fig. 1. A transverse section through the dorsal region of an embryo intermediate between Stages 1 and 11.
- Fig. 2-7. From a series of transverse sections through a younger embryo of Stage 11.
- Figs. 8-17. From a series of transverse sections through an older embryo of Stage 11.
- Figs. 18 and 19. Two sections from a series of cross-sections through a little more advanced embryo than the last.
- Figs. 20-29. From serial cross-sections through the most advanced embryo of Stage II.







## Plate XVIII.

a.pn.1-6, Anlagen of pronephric tubules from the first to sixth. a.sd., Anlage of segmental duct. cd., collecting duct. ch., chorda dorsalis. c.p., coelomic projection. cut., cutis-layer of myotome. fy., fore-gut. ep.. epiblast. hy.. hypoblast. l.m., lateral plate of mesoblast.

m.p., parietal layer of mesoblast.

ms., mesoblast.
mt.I. II. &c., the first, second, &c. myotome.
mus., muscle-layer of myotome.
m.v., visceral layer of mesoblast.
n., neural cord or canal.
pp.c., pleuroperitoneal cavity.
pt.1-6, pronephric tubules from the first to sixth.
sch., subchorda.
sd., segmental duct.

Figs. 30-31. From the same series as figs. 20-29 of the last plate.

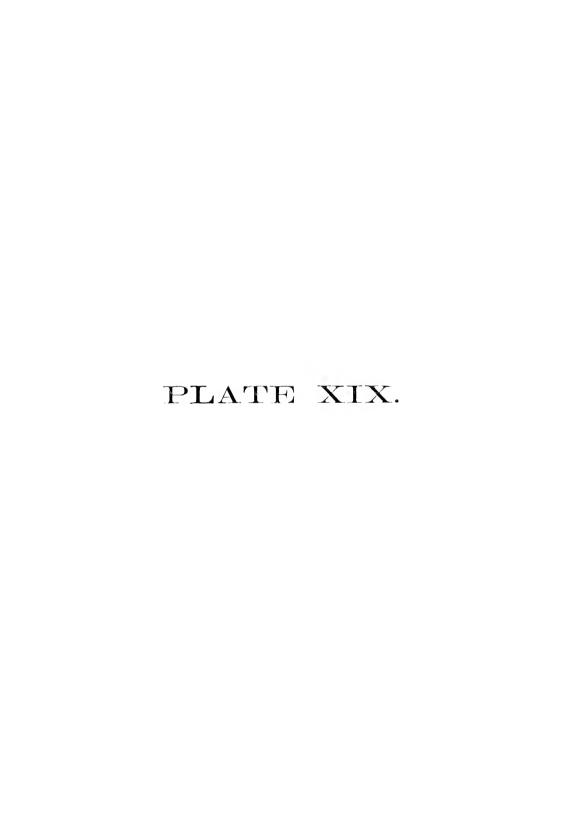
Figs. 32-50. From a series of transverse sections through a younger embryo of Stage III.

Figs. 51-58. From a series of transverse sections through an embryo of Stage III.

Fig. 59. A section through an older emboyo of Stage III, the posterior continuation of which is shown in the next following plate (figs. 60-63).





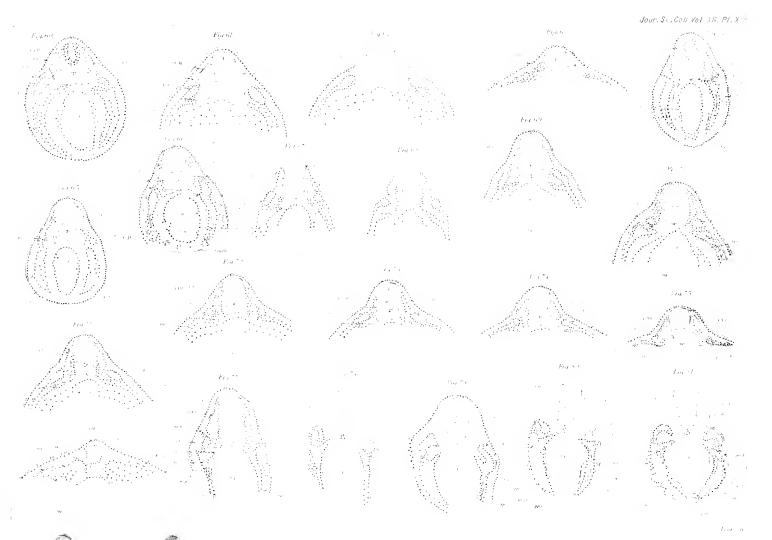


## Plate XIX.

cd., collecting duct.
ch., chorda dorsalis.
c.p., coelomic projection.
cut., cutis-layer of myotome.
d., dorsal row of mesoblast.
cp., epiblast.
fg., fore-gut.
l., Anlage of liver.
hy., hypoblast.
l.m., lateral plate of mesoblast.
mch., mesenchymatous cells.
ms., mesoblast.
m.p., parietal layer of mesoblast.

mt.I. II. &c., the first, second, &c. myotome.
mus., muscle-layer of myotome.
m.v., visceral layer of mesoblast.
n., neural canal.
nst.2-3, nephrostome the second and third.
pp.1-3, peritoneal partition.
pp.c., pleuroperitoneal cavity.
pt.1-6, pronephric tubules from the first to sixth.
sch., subchorda.
sd., segmental duct.
sy., spinal ganglion.
v. ventral row of mesoblast.

- Figs. 60-63. From the same series as, and the posterior continuation of, fig. 59.
- Figs. 64-76. From a series of transverse sections through an oldest embryo of Stage III.
- Figs. 77-81. From a series of transverse sections through an embryo of Stage IV; hence the body of embryo in the present stage is twisted, the sections pass through mavoidably oblique planes.





#### Plate XX.

u.sd., Anlage of segmental duct. bp., blastopore. brg., branchial region. cc., cloacal cavity. cd., collecting duct. ch., chorda dorsalis. c.dv. diverticulum of cloacal cavity. co.sd., cloacal opening of segmental duct. e.p., colomic projection. dl.bp., dorsal lip of blastopore. fg., fore-gut. ep., epiblast. gc. genital cells. hy., hypoblast. int., intestine. 7., Anlage of liver.

1.m., lateral plate of mesoblast. mch., mesenchymatous cells. ms.. mesoblast. m.p., parietal layer of mesoblast. mt.I, II, &c., the first, second, &c. myotome. m.r., visceral layer of mesoblast.  $n_{\cdot,\cdot}$  neural canal. nst.5, fifth nephrostome. perit., peritoncal membrane. pp.1-3, peritoneal partition.  $\rho \rho.c.$ , pleuroperitoneal cavity. pt.1-6, pronephric tubules from the first to sixth. sch., subchorda, sd., segmental duct. ye., volk-cells.

Figs. 82-89. Sections from the same series as fig. 81, lying posterior to it. The section shown in fig. 87 passes through somewhat frontally owing to the bending of the body-axis of the embryo; the neural canal, which is bent in the same manner as the axis meets with two time in section.

Wigs. 90 and 94. Two sections passing through in the same way as in fig. 87: in fig. 90 the dorsal lip of the blastopore, and in fig. 91, the upper (dorsal) portion of it, is cut through.

Figs. 92-96. From a series of transverse sections through a little older embryo than the last; the embryo is twisted in the same way as it.

Fig. 97. Frontal section through an embryo about the same stage as the last, the body of which has been straightened before cut through.

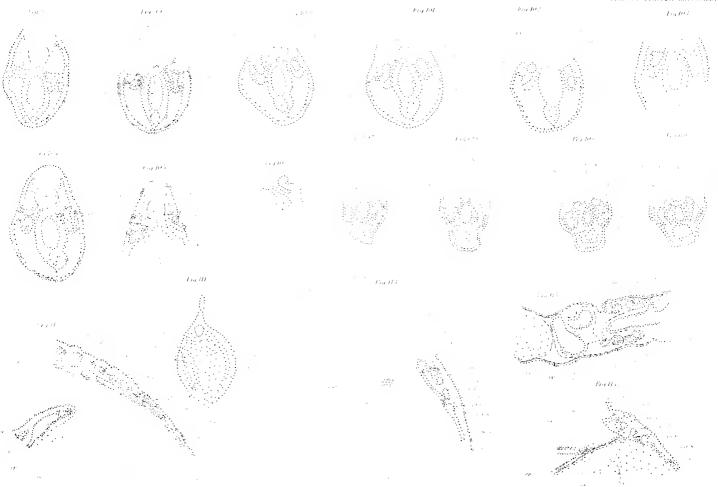




## Plate XXI.

1.m., lateral plate of mesoblast. a.cv., anterior cardinal vein. mch., mesenchymatous cells. au., auditory pit. m.p., parietal layer of mesoblast.  $b_{P}$ , ventral lip of blastopore. perit., peritoneal membrane. bry., branchial region. mt.I, II, &c., the first, second bs., blood space. cd., collecting duct. invotoine, &c. m.c., visceral layer of mesoblast. ch., chorda dorsalis. cc., cloacal cavity.  $n_{\bullet}$ , neural canal. c.dr., diverticulum of cloacal | nst.2-6, nephrostomes from the second to sixth. cavity. co.sd., cloacal opening of segpp.c., pleuroperitoneal cavity. mental duct. pt. 2- $\tilde{s}$ , pronephric tubules from cw., wall of cloaca. the first to fifth. ep., epiblast. r.m., radix of mesentery. sch., subchorda. ql., glomerulus of pronephros. sd., segmental duct. 7., dorsal fin. fg., fore-gut. t.a. tract of aorta. h., heart. t.ac. tract of anterior cardinal vein. hy., hypoblast. tr.a. truncus arteriosus. 7., liver, or Anlage of liver.

- Figs. 98-106. From a series of transverse sections through an embryo of Stage v.
- Figs. 107-110. From a series of transverse sections through an embryo of Stage vi.
- Fig. 111. Transverse section through the cloacal region of an older embryo of Stage vi.
- Figs. 112-114.  $\Lambda$  series of sagittal sections through a younger embryo in Stage iv.
- Fig. 115. A frontal section through an embryo a little more advanced than that of Stage vι.





# Beiträge zur Wachstumsgeschichte der Bambusgewächse.

Von

K. Shibata, Rigakushi.

Mit Tafeln XXII-XXIV.

# I. Einleitung.

Unsere Kenntnisse über Bau und Lebensweise der Bambusgewächse waren bisher sehr mangelhaft gewesen, obwohl in neueren Zeiten einzelne merkwürdige Erscheinungen auf dem Gebiete der Physiologie dieser eigenartigen Baumgräser durch interessante Beobachtungen¹) einiger die Tropen besuchender Botaniker zu Tage gefördert wurden. Bekanntlich gehören die meisten Bambuseen zu wärmeren Gegenden der alten und neuen Welt, mit Ausnahme von einigen kälterem Klima angepassten Formen, wie z. B. Bambusa Kurilensis, die in einer nördlichen Insel Japan's bei 46° n. B. gedeiht. Unser Land besitzt eine Reihe von Bambusformen, welche unserer Pflanzenphysiognomik ein charac-

<sup>1)</sup> G. Kraus, Physiologisches aus den Tropen. I. Längenwachstum der Bambusrohre. Ann. d. Jard, Bot. d. Buitenzorg. Vol. XII, p. 196.

H. Molisch, Über das Bluten tropischer Holzgewächse im Zustand völliger Belaubung. Ann. d. Jard. Bot. d. Buitenzorg. 1898. 2 tes Suppl. p. 23.

teristisches Aussehen verleihen. Einige hochwüchsige Formen aus der Gattung *Phyllostachys* sind bei uns überall häufig cultiviert, hauptsächlich für die mannigfaltigste Verwendbarkeit der Rohre und auch für ihre Frühjahrsschösslinge, die ein beliebtes Gemüse darbieten, während andere Arten aus *Arundinaria* und *Bambusa* als Zierpflanzen in unseren Gärten gemein sind.

Nun stellte ich es mir hier als Aufgabe, in erster Linie die Natur und das Verhalten der wichtigen Baustoffe während der verschiedenen Vegetationsperioden zu studieren, da mir besonders die ungemein rasche Entwicklung der Schösslinge etwas interessantes in Bezug auf Stoffwanderungs- und Stoffumwandlungsvorgänge darzubieten schien, oder in anderen Worten die Wachstumsgeschichte der Baumgräser mit Berücksichtigung der Bauverhältnisse näher zu verfolgen.

Die früheren Angaben über die Systematik, Verbreitung und äussere Lebensweise der Bambusgewächse haben eine Zusammenstellung in einem Werk Schröter's¹) erfahren, und es schien mir überflüssig dieselbe hier wiederzugeben. Was die Physiologie der Bambusgewächse anbetrifft, besitzen wir abgesehen von älteren Beobachtungen über das Wachstum der Schösslinge nur die eingangs erwähnten Arbeiten von Kraus und Molisch. Über die in Bambuspflanzen vorkommenden Stoffe besitzen wir ebenfalls spärliche Angaben. Cohn²) studierte "Tabaschir" in seiner klassischen Arbeit. Kozai³) stellte chemische Untersuchungen über die stickstoffhaltigen Bestandtheile des Schösslings von Phyllostachys mitis

<sup>1)</sup> C. Schröter, Der Bambus und seine Bedeutung als Nutzpflanze. Basel, 1895. Vergleiche ferner:

E. Hackel, Eambusacee. Engler's Die natürlichen Pflanzenfamilien. II, 2. p. 89. A. et C. Rivière, Les Bambous. Vegetation, culture et multiplication. 1878.

<sup>2)</sup> F. Cohn, Über Tabaschir. Beiträge z. Biol. d. Pflanzen. Bd. IV, p. 365.

<sup>3)</sup> Y. Kozai, On the nitrogenous non-albuminous Constituents of Bamboo shoots, Bulletin of the Cellege of Agriculture, Vol. I, Nr. 7.

an, und dabei fand er das Vorkommen von Tyrosin und Asparagin neben kleineren Mengen der Nucleinbasen. Die Angaben über die Bauverhältnisse der Bambusgewächse finden wir in Arbeiten von Schwendener, Haberlandt, Strasburger, Hohenauer, Güntz, Ross und Magnus. Näheres über die Litteratur wird noch an geeigneten Stellen Erwähnung finden.

Die vorliegenden Studien wurden im Laufe des academischen Jahres 1898-1899 im botanischen Institute der Kaiserlichen Universität zu Tokio ausgeführt. An dieser Stelle spreche ich meinem hochverehrten Lehrer Herrn Prof. Dr. Miyoshi meinem wärmsten Dank für seine vielfache Belehrung und Anregung aus. Es ist mir auch eine angenehme Pflicht Herrn Prof. Dr. Matsumura für seine gütige Unterstützung hier meinen herzlichsten Dank auszudrücken.

# II. Untersuchungsmaterial und Methodisches.

Als Untersuchungsobjecte dienten mir die im botanischen Garten der Universität cultivierten Bambusarten, insbesondere *Phyllostachys mitis*, Riv., die sich in dieser Gegend in voller Entwicklung findet und im hiesigen botanischen Garten auf ziemlich grossem Grund gepflanzt ist. Die Wachstumsgeschichte der Schösslinge der obengenannten Art wurde von der ersten Anlage bis zum mehrere Meter hohen Halmzustand verfolgt.

Auch die Schösslinge folgender Arten wurden zum Vergleichungszweck untersucht:

im April austreibende—Bambusa palmata, Bambusa Veitchii;
im Mai austreibende—Thyllostachys puberula, Arundinaria japonica;

im Juni austreibende—Phyllostachys bambusoides;

im Juli-August austreibende—Arundinaria Simoni, Arundinaria Hindsii;

im October - November austreibende—Arundinaria Matsumuræ, Arundinaria quadrangularis, Arundinaria Hindsii var graminca.

Die Entwicklung der Rhizomspitze wurde bei folgenden Arten im Herbst untersucht: *Phyllostachys mitis, Phyllostachys bambusoides*.

Für andere Arten, die ich in meiner Untersuchung gezogen habe, verweise ich auf das am Ende dieser Arbeit beigefügte Artenverzeichniss.

Um die Umwandlung und Wanderung der Stoffe in Reservestoffbehältern und in wachsenden Theilen zu verfolgen, bediente ich mich unter nöthigen Cautelen der üblichen microchemischen Methoden. Darüber sei hier folgendes bemerkt:

Stärke. Meyer'sche Chloralhydratjodlösung¹) wurde mit Vortheil benutzt.

Glycose. (Reducierender Zucker). Meyer'sche²) und Schimper'sche³) Methoden wurden neben einander angewandt, dabei hat sich die letztere zur Nachweisung der kleineren Menge geeigneter erwiesen. Obwohl diese üblichen Methoden auch zu unserem Zweck völlig ausreichten, habe ich noch Sicherheits wegen eine andere Reaction ausgeführt. Ich habe nämlich die Wasserauszüge von jungen Halmen, Wurzeln, Rhizomen und Scheideblättern und auch den Blutungssaft mit essigsaurem Phenylhydrazin erwärmt, und dabei erhielt ich stets characteristische gelbe Nadelkrystalle von Glucosazon.

<sup>1)</sup> Vergl. Strasburger, Botanisches Practicum. III. Auflage. p. 277.

<sup>2)</sup> A. Meyer, Microchemische Reaction zum Nachweis der reducirenden Zuckerarten-Ber, d. D. B. G. 1885. p. 332.

<sup>3)</sup> A. Zimmermann, Die botanische Microtechnik. p. 75.

Rohrzucker. Die Unzuverlässigkeit der bekannten Sachs'schen Methode wurde vielfach von Autoren betont. Ich habe nur ausnahmsweise diese Reaction benutzt, während in den meisten Fällen ich mich der neulich von Hoffmeister<sup>1</sup>) aufgestellten Invertinmethode bediente.<sup>2</sup>)

Gerbstoffe. Die Eisensalzlösungen, insbesondere die ätherische Lösung des Eisenchlorids, und auch die Kaliumbichromatlösung wurden angewandt.

Fette. Alkannatinctur und 1-procentige Osmiumsäurelösung wurden benutzt.

Asparagin. Die bekannte Borodin'sche Methode<sup>3</sup>) hat sich zweckentsprechend erwiesen. Zur Erkennung der erhaltenen Krystalle als Asparagin diente mir hauptsächlich die Winkelmessung. Vielfach wurde das Borodin'sche Verfahren mit gesättigter Asparaginlösung angewandt. Ferner diente mir Diphenylamin-Schwefelsäure zur Unterscheidung von Asparagin und Salpeter.

Tyrosin. Die nach Borodin'scher Methode behandelten Schnitte ergaben eine reichliche Ausscheidung von eigenthümlichen Krystallen, die ohne Schwierigkeit mit Tyrosin identificiert

<sup>1)</sup> C. Hoffmeister, Über den microchemischen Nachweis von Rohrzucker in pflanzlichen Geweben. Jahrb. f. wiss. Bot. Bd. XXXI, p. 688.

<sup>2)</sup> Die von den "Ebisu"-Brauerei bezogene Hefe-Reinkultur wurde zur Darstellung von Invertin verwendet, dabei habe ich wie folgt verfahren: Die Hefemasse wurde mittelst Filtration von der Kulturflüssigkeit befreit und nach wiederholtem Auswaschen mit Wasser zum dicken Brei angerührt. Der Hefebrei kam nach dem Verreiben im Mörser in den Thermostat bei 50°C, in welchem er für 10-12 Stunden gelassen wurde. Hiernach wurde die abfiltrierte Flüssigkeit mit 90% Alcohol versetzt, und der dabei entstandene voluminöse Niederschlag auf Filtrirpapier gesammelt, welcher, nach wiederholtem Auswaschen mit 90% Alcohol und dann mit absolutem Alcohol auf Schwefelsäure getrocknet wurde. Die wässrige Lösung der erhaltenen weissen kreideartigen Substanz, die allein niemals Fehling' sche Lösung reduciert, zeigte ein energisches Inversionsvermögen. Bei wiederholten Versuchen habe ich ferner in keinem fall die Beimengung von diastatischen und cellulosespaltenden Enzymen gefunden. Weitere Verfahren mit den Schnitten genau nach Hoffmeister.

<sup>3)</sup> A. Zimmermann, Die botanische Microtechnik. p. 80.

wurden') (Fig. 58). Belzung'sche Glycerin-Methode<sup>2</sup>) wurde auch mit Vortheil benutzt, wobei sich schöne Nadelbüschel in den Zellen bilden (Fig. 61). Ich habe ferner zur Erkennung der Vertheilung des Tyrosins in eiweissarmen Gewebetheilen Millon's Reagens benutzt, und dabei wurden die tyrosinreichen Zellen schnell blutroth gefärbt. Die Färbung bleibt nach dem Auslaugen der zuvor mit absolutem Alcohol behandelten Schnitte mit dem warmen Wasser für 10-20 Minuten so gut wie gänzlich aus. Daher kann diese Rothfärbung niemals von Eiweissstoffen herrühren.

Eiweiss Biuretreaction und Raspail's Reaction wurden vornehmlich benutzt. Millon's Reagens kam zur Anwendung erst nach dem Ausziehen von Tyrosin in oben beschriebener Weise.

Mineralstoffe. Die von Schimper") empfohlenen Reagentien wurden verwendet. Die Controllversuche wurden öfters ausge-

<sup>1)</sup> Dies geschah aus folgenden Gründen:

Die Gestalt der Krystalte. Die feine Nadelbüschel in dendritischer Gestalt oder Doppelpinselform bietet ganz dasselbe Aussehen wie reines Tyrosin.

Das optische Verhalten. Im durchfallenden Licht erscheinen die Krystalle bräunlich und im auffallenden Licht weisslich seidenglänzend. Im polarisierten Licht zeigen die Krystalle starke Doppelbrechung.

<sup>3.</sup> Die Löslichkeitsverhältnisse. Die Krystalle sind unföslich in kaltem Wasser, aber löslich in kochendem Wasser, Ammoniak und verdünnter Salzsäure. Ferner sind die Krystalle unföslich in heissgesättigter Tyrosinfösung.

<sup>4.</sup> Das Verhalten beim Erhitzen. Wenn man den mit Tyrosinkrystallen besetzten Objectträger auf der Flamme erhitzt, bis nebenbei vorhandene Asparaginkrystalle sich zu braunen Schäumen verwandeln (ca. 200° C), so sieht man, dass die Nadelkrystalle ganz unverändert bleiben.

Das Verhalten gegen Millon's Reagens. Die Krystalle l\u00fcsen sich im Millon's Reagens mit einer prachtvoll rothen F\u00e4rbung der umgebenden Fl\u00fcssigkeit.

Die oben angeführten Merkmale reichen aus, die Krystalle microchemisch als Tyrosin zu erkennen.

Belzung, Recherches chimique sur la Germination. Ann. d. Sc. nat. Bot. Sér. VII. T. 15, p. 209.

<sup>3)</sup> A. F. W. Schimper, Zur Frage der Assimilation der Mineralsalze durch die grüne Pflanze. Flora. Bd. 73, 1890, p. 210.

führt, um die Reinheit der Reagentien zu prüfen. Die microchemischen Reactionen wurden sowohl an frischen Schnitten als an auf Deckgläsern geglühten Aschen vorgenommen.

#### III. Die Bauverhältnisse.

Die Bauverhältnisse der Bambusen sind bisher spärlich und nur gelegentlich Gegenstand der anatomischen Forschung geworden. Strasburger¹) hat den Bau des Gefässbündels von Bambusa vulgaris kurz geschildert. Das mächtig entwickelte Bastgewebe in Bambushalmen wurde vielfach von Schwendener²) Haberlandt³) u. A. erwähnt. Ross⁴) bemerkte den anomalen Bau der Wurzeln. Die Betrachtungen über die Blattstructur finden wir in den Arbeiten von Güntz³), Magnus⁶), Haberlandt³) und Schwendener⁶). Ubrigens liegen uns noch einige kurze Angaben von Hohenauer⁶) und Möbius¹⁰) vor. Nun schien es mir wünschenswerth die Bauverhältnisse der Vegetationsorgane der Bambusgewächse einem genaueren Studium zu unterwerfen, damit für die physiologische Forschung dieser interessanten Pflanzengruppe eine festere Grundlage geschaffen werde. Meine

Nr. 51, p. 479).

<sup>1)</sup> Strasburger, Über d. Ban u. Verrichtungen d. Leitungsbahnen. 1891. p. 363.

<sup>2)</sup> Schwendener, Das mechanische Princip in anat. Bau d. Monocotylen. p. 65.

<sup>3)</sup> Haberlandt, Entwicklungsgeschichte des mech. Gewebesystems d. Pflanzen. p. 23.

<sup>4)</sup> Ross, Beiträge z. Anatomie d. abnorm. Monocotylenwurzeln. Ber. d. Deutsch. Bot. Gesells, Ed. I, p. 338.

<sup>5)</sup> Güntz, Unters. üb. d. anat. Structur d. Gramineenblätter. p. 37, 41, 44, 48, 64 etc.

<sup>6)</sup> Magnus, Einfalzungen d. Zellmembran. (Just's Jahresber. d. Bot. I, p. 367).

<sup>7)</sup> Ha'berlandst, Vergl. Anat. d. jassim. Gewebesystems d. Pflanzen. Jahrb. f. wiss. Bot. Bd. XIII, p. 100.

<sup>8)</sup> Schwendener, Die Mestomscheide der Gramineenblätter. Ges. Bot. Mitt. Bd. II, p. 178.

<sup>9)</sup> Hohenauer, Vergl. anat. Unters. üb. d. Bau d. Stammes bei d. Gramineen. p. 556. 10) Möbius, Üb. d. eigent. Blühen von Bambuså vulgaris. (Ref. in Bot. Centralbl. 1899.

diesbezügliche Untersuchungen erstreckten sich auf sieben und zwanzig vorwiegend einheimische Formen, die sich in die vier Gattungen von *Phyllostachys*, *Arundinaria*, *Bambusa* und *Dendrocalamus* vertheilen. Die wesentlichen Ergebnisse will ich in folgenden Zeilen kurz darzustellen versuchen.

### Das Rhizom.

Die Rhizome<sup>1</sup>) von *Phyllostachys*- und *Arundinaria*-Arten sind bekanntlich kurz gegliederte horizontal verlaufende Stengelgebilde mit einer rundlichen oder rundlich-ovalen Querschnittform. Die internodiale Markhöhle ist stets stark reduciert, meist nur einige mm breit und kommt nicht selten zum gänzlichen Verschwinden. Wir wollen zunächst beispielweise ein Rhizominternodium von *Phyllostachys mitis* ins Auge fassen.

Nächst unter der Epidermis kommt ein 1-3 schichtiger Ring von den englumigen langgestreckten sklerotischen Parenchymzellen, deren Querwände öfters etwas schief gestellt sind. Das darinnen liegende 20-25 schichtige bündelfreie Parenchym wird als die primäre Rinde<sup>2</sup>) aufgefasst. Es geht ohne scharfe Grenze zum Grundgewebe des Centralcylinders über, in welchem in üblicher Weise die collateral gebauten Gefässbündel zerstreut liegen. Sämmtliche Parenchymzellen sind verholzt und mit zahlreichen ovalen Tüpfeln versehen. In diesem Gefässbündel erblickt man ein typisch gebautes Gramineenbündel. Die grosse Lumenweite der Siebröhren ist dabei höchst auffallend<sup>3</sup>); es wurde oftmals einen Durchmesser von 0.15 mm erreicht<sup>4</sup>), während

<sup>1)</sup> Vergl. A. et C. Rivière, Les Bambous, p. 68, p. 236.

<sup>2)</sup> Falkenberg, Vergl. Unters. üb. d. Vegetationsorg. d. Monocotylen. p. 163.

<sup>3)</sup> Strasburger, Leitungsbahnen. p. 363.

<sup>4)</sup> Die Angaben über die Lumenweite der Siebröhren einiger Pflanzen findet man bei Lecomte (Ann. d. Sc. nat. Bot. Ser. VII, T. X, p. 242).

die grösste Parenchymzelle 0.09 mm und die Geleitzellen meist nur 0.005 mm weit sind. Die beiden seitlichen Gefässe, deren maximale Lumenweite 0.2-0.3 mm beträgt, communicieren mit den einschichtigen Belegzellen durch regelmässig angereihte quergestreckte Tüpfel. Die stark entwickelten Bastbelege um die Gefässbündel sind seitlich an der Grenze zwischen Hadrom und Leptom und oft auch unterhalb der seitlichen Gefässe unterbrochen. Dadurch kommen ein oder zwei Paar Durchlassstellen zu Stande, die, wie zuerst Schwendener¹) bemerkt hat, einen Stoffaustausch zwischen Bündelelementen und Grundgewebe ermöglichen.

Wenn man die Querschnittbilder der Rhizominternodien der übrigen Arten in Betracht zieht, so lassen sich unter denselben folgende drei Typen unterscheiden, nämlich:

Erster Typus. Die äussersten Bündel, welche direct an die Rinde grenzen, stehen vollkommen isoliert von einander. Hierher gehören Phyllostachys mitis, Phyllostachys bambusoides, Phyllostachys puberula und Arundinaria Narihira.

Zweiter Typus. Die Bastbelege der äussersten Bündel versehmelzen sich häufig unter einander und auch mit den Baststrängen zu unregelmässigen Bastbändern. Als Beispiele dienen Arundinaria japonica, Bambusa borealis, Arundinaria Tootsik, A. Simoni, A. Hindsii etc. In den zwei letztgenannten Arten befindet sieh jedoch oft ein nahezu vollkommener Bastring. (Fig. 3).

Dritter Typus. Der echte subcorticale Bastring<sup>2</sup>), an welchen die Mestombündel innen angelehnt sind, befindet sich bei

<sup>1)</sup> Schwendener, Das mechanische Princip. p. 107.

<sup>2)</sup> Vergl. Haberlandt, Entwicklungsgeschichte des mechan. Gewebesystems p. 28.; Physiologische Pflanzenanatomie. p. 157.

Bambusa palmata, B. Veitchii, B. paniculata, B. nipponica, B. ramosa, B. nana, Arundinaria quadrangularis, A. Matsumuræ, A. variabilis, Arundinaria pygmæa und ferner Phyllostachys Kumasasa. (Fig. 2).

Der Bastring der letzterwähnten Arten, welcher je nach Species verschieden stark ausgebildet ist, geht entwicklungsgeschichtlich aus einem entsprechend continuirlichen Cambiumring¹) hervor. Schwendener sagt²): "Für Bambuseen ist die Querschnittform des mechanischen Systems, wie ich sie früher beschrieben habe, characteristisch genug, um jede nähere Verwandtschaft mit den Festucaceen oder irgend einem anderen Tribus auszuschliessen. Ein Bastring ist nicht vorhanden,......" Diese Bemerkung Schwendener's passt aber nach obigem Befunde auf die Rhizome nicht.³) Der subepidermale Sklerenchymring ist nur schwach entwickelt, es ist bei den meisten Arten nur 1-2 Schichten dick. Die Dicke der primären Rinde ist meist unanschnlich und variirt zwischen 4-35 Zellschichten.

Der Bastring wird stets vielfach unterbrochen in den Knoten, um hier den neu eintretenden Blattspursträngen Platz zu machen. Ferner ist es als die Regel hervorzuheben, dass innerhalb des Knotens der Bastbeleg des Mestombündels eine bedeutende Reduction erfährt, und sich meist nur auf eine dünne Sichel um das Leptom beschränkt. Die sämmtlichen Elemente des Bündels sind hier kurzgliedrig, und die Seitenwände der Siebröhren sind mit den ausserordentlich zahlreichen Siebtüpfeln versehen. Bekannt-

<sup>1)</sup> Haberlandt, Entwicklungsgeschichte. p. 28.

<sup>2)</sup> Schwendener, Die Mestomscheide der Gramineenblätter. p. 183.

<sup>3)</sup> Die mittleren Durchmesser der Rhizome dieser drei Typen stehen ungefähr im Verhältnisse 6:3:1. Die Ausbildung der Bastplatte resp. des Bastrings in Rhizomen entspricht wohl den mit der Dünnheit steigenden Aufforderungen für die Biegungsfestigkeit. Jedenfalls gehört hier die Anordnung des mechanischen Systems in Rhizomen nicht zum sogenannten taxonomischen Merkmalen.

lich findet sieh in dem Knoten die Vereinigung der Blattspurstränge unter einander und mit den Achselknospenbündeln statt<sup>1</sup>). Die hier eintretenden Knospenbündel verbreiten sich in dem Knotengewebe nach allen Richtungen hin. Die Gefässe der Knospenbündel setzen sich in üblicher Weise unter starker Krümmung an die der Blattspuren an2). Dennoch verdient die Art und Weise, wie der Übergang des Leptoms erfolgt, eine besondere Beachtung. Das Leptom des Knospenbündels ist bei der Ansatzstelle an Blattspuren so stark angeschwollen, dass ihr ganzer Umriss mit einer Spindel zu vergleichen ist. Figur 4 stellt ein derartiges Gebilde dar. Diese angeschwollene Partie weicht von dem üblichen Bau des Leptoms in so fern ab, dass sie die Differenzierung ihrer Elemente in Siebröhren und Geleitzellen nicht mehr aufweist, sondern aus lauter gleichartigen feinen ca. 5-6 µ breiten cambiformartigen Elementen<sup>3</sup>) zusammengesetzt ist (Fig. 4 u. 8), und folglich im Querschnittbild ein regelmässiges englumiges Maschenwerk darstellt (Fig. 5 u. 7). Die Anordnung dieser feinen cambiformartigen Elemente bietet eine grosse Eigenthümlichkeit dar. Die etwas schräg gestellten Endflächen der seitlich an einander stossenden Elemente scheinen ungefähr auf derselben Querebene zu liegen, und demgemäss stellt der ganze spindelförmige Theil im Längsschnitt ein der Länge nach an einander angereihtes meist 5-7 faches Stockwerk von cambiformartigen Elementen dar. Die Elemente in den mittleren 1

<sup>1)</sup> Vergl. Falkenberg, Vergl. Unters. d. Vegetationsorgane. p. 187.

<sup>2)</sup> Strasburger, Leitungsbahnen. p. 353 u. p. 365.

<sup>3)</sup> Selbst bei den relativ weiteren (z. B. bei Arundinaria Simoni) überschreitet die Breite kaum 9 v. So weit ich unterrichtet bin, wurde derartige Structur bisher in keiner anderen Pflanzen beobachtet. Zum Beispiel finden wir keine diesbezügliche Angabe bei verschiedenen Monocotylen, die von Falkenberg (loc. cit.) und Strasburger (loc. cit.) gründlich untersucht wurden. Ob sie auch bei anderen Pflanzen vorkommt muss deshalb zur Zeit dahingestellt bleiben.

oder 2 Etagen dieses Stockwerks sind sehr langgestreckt und besitzen fein undulierte Seitenwände mit zahlreichen grossen ovalen tüpfelartig verdünnten Stellen, die zum Beispiel bei Phyllostachys mitis eine Weite von  $5 \times 3$   $\mu$  erreichen (Fig. 6). In einem Ende dieses spindelförmigen Theils vermitteln die etwas breiteren Elemente,—die sich zu 4-5 je einer feinbetüpfelten Endfläche der Siebröhren und einzeln auch den Geleitzellen anschlissen,—den Uebergang zum normal gebauten Leptom des Knospenbündels (Fig. 10). Das andere Ende der Spindel setzt sich in verschiedener Neigung und oft sogar rechtwinkelig an das Leptom der Blattspuren an (Fig. 8). Einige Schichten der Scheideelemente grenzen gewöhnlich den spindelförmigen Theil Die Zellwandbeschaffenheit der vom Grundparenchym ab. spindelartigen Theile weicht kaum von der des Leptoms ab; sie zeigen nämlich ebenso starke Cellulose-Reaction mit Chlorzinkjod oder Schwefelsäure-Jod, und sie werden auch mit Anilinblau, Congoroth u.a., im nahezu gleichen Farbenton wie Siebröhren gefärbt. In späterem Alter tritt jedoch oft eine Spur Holzreaction in den Elementen der oben erwähnten mittleren Etagen ein. Was den Plasmagehalt dieser Theile anbetrifft, so scheint es nur auf einen zarten Wandbeleg beschränkt zu sein, wie es bei Siebröhren stets der Fall ist. In den ersten Entwicklungszuständen habe ich constatiert, dass diese Anschwellung aus den entsprechend vermehrten Längstheilungen der procambialen Zellen an der betreffenden Stelle hervorgeht. In solchen früheren Stadien zeichnet sich diese Anschwellung durch besonders reichlichen Plasmagehalt und auffallend grosse Zellkerne aus, wie es Fig. 9 zeigt.

Derartige spindelförmige Leptomanschwellungen in Knospenbündeln habe ich regelmässig in Rhizomknoten sämmtlicher von mir untersuchter Arten gefunden, aber man findet sie am stärksten ausgebildet in den Rhizomknoten der Phyllostachys-Arten, wobei ihre Querschnittgrösse sogar einem grossen Mestombündel nahekommt. Da die an Rhizombündel sich ausetzenden Achselknospenstränge in ihrer Gesammtheit ein physiologisches Analogon des haustorial Saugorgans darstellen, so ist es nicht unmöglich, dass dieses Gebilde in dem Sinne ausgebildet ist, dass es eine specifisch absorbierende Wirkung auf die Siebröhren der Mutterrhizome auszuüben vermag. Allerdings besitzt es in seinen anatomischen Merkmalen vieles gemein mit den üblichen Absorptionsgeweben<sup>1</sup>). So lange aber die Stofftransportmechanik im Leptome noch nicht in allen Hinsichten aufgeklärt ist<sup>2</sup>), möge die nähere Erörterung der physiologischen Vorgänge, die sich in diesem abweichend gebauten Leptomtheile abspielen, auf eine künftige Gelegenheit verschoben werden.

### DER HALM.

Die Bambushalme<sup>3</sup>) sind bekanntlich mit den hohlen Internodien versehen, die bei *Phyllostachys mitis* oft eine ansehnliche Dicke von 20 cm erreichen.

Die primäre Rinde ist stets weit schwächer entwickelt als in dem Rhizome; diese Verhältnisse, wie sie schon von Falkenberg<sup>4</sup>) und Rothert<sup>5</sup>) für andere Pflanzen nachgewiesen wurden, gehen noch aus den folgenden Beispielen deutlich hervor:

<sup>1)</sup> Haberlandt, Physiologische Pflanzenanatomie. p. 186.

<sup>2)</sup> Czapek, Über d. Leitungswege d. organischen Baustoffe in Pflauzenkörper. p. 24; Lecomte, Etude du Liber des Angiospermes. Ann. d. Sc. nat. Ser. VII. T. X, p. 303.

<sup>3)</sup> Vergl. Rivière, Les Bambous. p. 134.

<sup>4)</sup> Falkenberg, l.c. p. 134.

<sup>5)</sup> Rothert, Vergl. anat. Unters. üb. d. Differenzen im prim. Bau d. Stengel u. Rhizome. p. 92.

	1	HALM			HIZOM	
	Durchmesser des Central- cylinders	Dicke der Rinde	Q®	Durchmesser des Central- cylinders	Dicke der Rinde	Q®
Phyllostachys mitis	130.0	0.315	412	23.8	0.728	33
Phyllostachys bambusoides	34.0	0.059	575	20.0	0.611	33
Phyllostachys puberula	56.0	0.049	1142	22.0	0.933	23
Phyllostachys Kumasasa	2.7	0.023	120	5.6	0.780	7
Arundinaria Simoni	14.0	0.049	285	8.0	0.494	16
Arundinaria japonica	17.0	0.050	340	8.4	0.286	29
Arundinaria Hindsii	22.0	0.059	373	18.0	0.364	49
Arundinaria quadrangularis	23.0	0.045	511	8.0	0.260	31
Arundinaria Matsumuræ	2.5	0.018	139	2.9	0. 20	15
Arundinaria pygmæa	2.3	0.036	64	4.5	0.325	14
Arundinaria Narihira	14.0	0.049	285	21.0	0.468	45
Bambusa borealis	5.5	0.045	122	6.5	0.212	31
Bambusa palmata	10.0	0.063	159	7.1	0.624	11
Bambusa Veitchii	4.5	0.023	200	2.9	0.143	20

\*Q.=Das Verhältniss des ersteren zur letzteren.

Die äussersten 1-2 Schichten Rindenparenchymzellen sind oft sklerotisch verdickt (Fig. 12) und unterscheiden sich von den übrigen nur durch eine grössere Länge; folglich haben wir hierbei keineswegs mit einem Bastring, wie Haberlandt einst annahm,¹) zu thun. Die Bastbelege der peripherischen Gefässbündel und die dazwischen liegenden Baststränge verschmelzen mit einander zu unregelmässigen Bastbändern, besonders häufig in dünneren Halmen von Arundinaria Matsumuræ, A. pygmæa etc. Dennoch begegnet man hier in keinem Fall dem echten Bastringe.

<sup>1)</sup> Haberlandt, Entwicklungsgeschichte. p. 23.

Die zuerst von Schwendener¹) bei einigen Bambusaarten entdeckte eigenthümliche Parenchymlamelle, die quer in dem innenseitigen Bastbelege inseriert ist, habe ich auch in den Halmen aller echter Bambusaarten (B. vulgaris, B. nana und B. stenostachya), Dendrocalamus latiflorus und bei 2 Arundinariaarten (A. Hindsii, A. quadrangularis) aufgefunden. Überdies habe ich die Fälle beobachtet, dass die Lamelle nur an einer Seite in das Grundparenchym übergeht, und dass sogar das Parenchym in der Mitte des Beleges allseitig von Bastzellen umschlossen liegt (Fig. 17 u. 18). Nach den letzterwähnten Thatsachen erscheint es a priori sehr wahrscheinlich, dass dieses Parenchymgewebe erst nachträglich aus einem Theil des Procambiums des Bastbeleges hervorgeht, wie es Haberlandt²) schon vermuthet hat.

In der That konnte ich in einem Procambialstrang in jungen Internodien zuerst nichts von dieser Parenchymlamelle erkennen. Sie differenziert sich erst später aus einer Stranganlage, in welcher alle Formelemente schon fertig angelegt sind, derart, dass die langgestreckten Procambialzellen in betreffender Stelle successive Quertheilungen erfahren und zum Epen umgewandelt werden (Fig. 21 u. 22). Die feinkörnige Stärke, die später dem Zucker Platz macht, tritt sogleich in diesem Gewebe auf (fig. 20 u. 22) und bleibt in demselben während der weiteren Ausbildung des Stranggewebes. In dieser Weise dient die Parenchymlamelle den dem Mestom unmittelbar anliegenden Bastzellen als ein Speicherungsort der nötigen Baustoffe. Die durch diese Parenchymlamelle vom Mestom abgetrennte Bastmasse bleibt gewöhnlich in ihrer Ausbildung sehr zurück, wie das Fig. 19 zeigt. Nun schien es mir berechtigt diese Paren-

<sup>1)</sup> Schwendener, Das mechanische Princip. p. 65.

<sup>2)</sup> Haberlandt, l.c. p. 23.

chymlamelle als eine im Innern des Stranggewebes eingeschobene "Stärkescheide"), die in ihrer physiologischen Rolle der gewöhnlichen strangumgebenden gleicht, aufzufassen. Derartige Einrichtungen würden vielleicht zweckentsprechend sein, bei einem mit so starkem Bastbelege versehenen Bündel, wie es bei Bambuseen angetroffen wird²).

Die schon erwähnten spindelförmigen Leptomanschwellungen des Knospenbündels kommen auch in dem Halmknoten vor, jedoch meist in schwächerer Ausbildung.

Die dünneren Blatttragenden Zweige stimmen in ihrem Bau mit den dickeren Halmtheilen wesentlich überein. Bei einem solchen (mit einem Durchmesser kleiner als 1 mm) wird das Rindenparenchym zu 1-2 Schichten reduciert und mehr oder minder verdickt. Die aussenseitigen Bastbelege der peripherischen Bündel stossen oft direct an die Epidermis, so dass eine Art Bastrippe zu Stande kommt<sup>3</sup>). Derartige Rippenbildung konnte ich jedoch bei einigen Arten, wie Arundinaria Matsumuræ, selbst in den dünnsten Zweigen (0.7 mm dick) nicht nachweisen. Was den Gefässbündelverlauf in den Halmen sowie in den Rhizomen anbetrifft, so gehört er dem Palmentypus<sup>4</sup>) an, and habe ich durch successive Querschnitte und Längsschnitte in der Spitzenregion constatiert, dass die grossen medianen Blattspurstränge 5-6 Internodien zurücklegen müssen, bevor sie sich an andere Blattspurstränge ansetzen.

<sup>1)</sup> Vergl. Heine, Über physiologische Function der Stürkescheide. Ber. d. D. B. G. 1885, p. 189.

<sup>2)</sup> Allerdings wurde die mechanische Bedeutung, die Detlefsen (Üb. d. Biegungselasticität v. Pflanzentheilen. Arb. d. Bot. Inst. Würzburg. Bd. III, p. 182.) diesen Parenehymlamellen zuzuschreiben versuchte, von Schwendener (Zur Lehre v. d. Festigkeit d. Gewächse. Ges. Bot. Mitteil. Bd. II. p. 19-20.) genügend widerlegt.

<sup>3)</sup> Vergl. Schwendener, Die Mestomscheide. p. 183.

<sup>4)</sup> De Bary, Vergleichende Anatomie d. Vegetationsorgane. p. 271 ff.

### DER STIEL.

Die vielen untersten Internodien des Schösslings von *Phyllostachys mitis* vereinigen sich sehr früh zu einem verholzten soliden Gebilde, welches im fertigen Zustande 2-4 cm lang und nur 1.0-1.5 cm dick ist. Das Gebilde, das ich hier "Stiel" nenne, lässt keinen Unterschied mehr zwischen Internodien und Nodien im inneren Bau erkennen.

Die Rinde dieses Theils besteht aus etwa 20 Schichten parenchymatischer Zellen. Die Bastbelege der äussersten Bündel verschmelzen sich zu einem vielfach unterbrochenen unregelmässigen Band. Nach innen liegen zahlreiche Bündel, welche die ganze Länge des Stiels hindurch gedrängt verlaufen (Fig. 13) und dann in Rhizomknoten eintreten, um sich dort an die Blattspuren anzusetzen. So überwiegen im Querschnitte des Stiels sehr stark die Bündel, und das dazwischen liegende Parenchym ist zu einem 2-4 schichtigen schmalen Gewebe reduciert. Diese Verhältnisse entsprechen wohl der Function des Stiels als Leitungswege und nicht als Speicherngsort. Die Querschnittform des Bündels mit Bastbelege ist in den meisten Fällen rundlich oval und es ist vollkommen von 3-6 schichtigen Bastzellen umgeben. Daher ist der Stoffaustausch zwischen leitenden Elementen und Grundparenchym so gut wie gänzlich ausgeschlossen. Das Leptom nimmt die äussere Hälfte des Bündels ein und besteht aus einer Anzahl 0.04-0.05 mm breiten Siebröhren und englumigen Geleitzellen. Das Hadrom besteht aus nur einem (seltener zwei) grossen Gefässe (oft bis 0.15 mm weit), welches von einigen Schichten kleinzelligen Hadromparenchyms

<sup>1)</sup> Dieser "Stiel" stellt also eine einzige Stoffleitungsbahn zwischen dem vom Schösslinge 5'ch entwickelnden Halme und dem Rhizome dar.

umgeben ist (Fig. 14). Dazu kommen noch einige Tracheiden mit netz-, spiral- oder ringförmigen Wandverdickungen. Bei den übrigen Arten sind die ebenso schmalen Stieltheile in genau derselben Weise ausgebildet und sie stimmen in ihrer inneren Structur mit dem oben beschriebenen gänzlich überein.

### DIE WURZEL.

Die zahlreichen Wurzeln¹) befinden sich radial angeordnet an den Rhizomknoten und den unterirdischen Halmknoten; sie erreichen bei *Phyllostachys*-Arten eine maximale Länge von 70 cm mit einem Durchmesser von 4 mm. Zunächst will ich hier den anatomischen Ban der Wurzelrinde von *Phyllostachys*- und *Arundinaria*-Arten näher betrachten.

Die äusserste Zellschicht der Rinde lässt sich als die Aussenscheide unterscheiden, indem die äusseren und radiaren Zellwände sehr stark verdickt sind (Fig. 24a u. 25). Damit spielt sie die Rolle der schützenden Oberhaut anstatt der Epidermis, die sehr früh zerstört und abgeworfen wird. Nach innen folgt die verholzte Bastschicht (Fig. 24a). Das Rindenparenchym lässt sich in die äusseren aus unregelmässig polygonalen weitlumigen Zellen zusammengesetzten Schichten und die inneren aus regelmässig in radialen und concentrischen Reihen angeordneten Zellen bestehenden Schichten unterscheiden<sup>2</sup>). Die letzteren sind von einer Anzahl radialer Lufträume durchzogen. Die Zellen der Endodermis besitzen eine starke Verdickung von inneren und radialen Wänden, die in üblicher Weise verkorkt sind (Fig. 28). Die stark verdickte Wandung ist zierlich ge-

<sup>1)</sup> A. et C. Riviere, Les Bambous, p. 93.

<sup>2)</sup> Die Zellschichtenzahl der inneren Rinde ist stets kleiner als die der äusseren.

schichtet und von feinen verästelten Kanälen förmlich durchsetzt (Fig. 28). Im Querschnitt stellt demnach diese nach Aussen zugekehrte C-Scheide ein symmetrisches Bild mit der oben erwähnten ebenso C-förmigen Aussenscheide dar<sup>1</sup>). Die 1 oder 2 innersten unmittelbar der Endodermis anliegenden Rindenschichten sind bei *Phyllostachys Kumasasa*, *Bambusa borealis* und *Arundinaria quadrangularis* als Verstärkungsring<sup>2</sup>) ausgebildet, indem die Zellen durch innenseitige C-förmig verdickte und stark verholzte Wände ausgezeichnet sind (Fig. 34).

So weit es den Bau der Wurzelrinde betrifft, zeigen die echten Bambusa-Arten nämlich B. vulgaris, B. nana, B. stenostachya, B. arundinacea u. a. ein von dem oben beschriebenen ganz abweichendes Verhalten. Hier weisen die Zellen der subepidermalschicht keine Wandverdickung auf und ebenso verhalten sieh die persistenten Epidermiszellen. Darauf folgen 2-3 Schichten enger Bastelemente, welche nach innen scharf von den weitlumigen Rindenparenchymzellen abgesetzt werden (Fig. 26 u. 27). Die äussere Rinde besteht nur aus einigen Zellschichten, während die von grossen Lufträumen durchzogenen inneren Schichten vielfach dicker sind (Fig. 27). Die Endodermiszellen sind ringsum verdickt und bilden die sogenannte O-Scheide3). Merkwürdig ist ferner der Bau des Verstärkungsrings. Die innersten 1-oder 2-schichtigen Rindenparenchymzellen führen an ihren inneren Wänden eine Anzahl unregelmässig gestalteter aus reiner Cellulose bestehender Auswüchse, die häufig die äusseren Wände erreichen, so dass sie im Querschnitt die ganzen Zellen nahezu aus-

<sup>1)</sup> Dasselbe Verhältniss wurde von Schwendener (Die Schützscheiden und ihre Verstärkungen, Ges. Bot. Mitt. Bd. II, p. 120, 127.) auch bei einigen Orchideenluftwurzeln bewegt:

<sup>2)</sup> Sehwendener, Die Schützscheide und ihre Verstärkungen. Ges. Bot. Mitt. p. 132.

<sup>3)</sup> Vergl. Schwendener, l.c. p. 128, Tabelle.

zufüllen scheinen (Fig. 31 u. 32)¹). Eine Anzahl einheimischer Arten, die wegen ihrer 6 Stamen bisher in Bambusa eingereiht wurden, z. B. B. Veitehii, B. palmata, B. borealis etc., weisen jedoch im Bau der Wurzelrinde eine vollkommene Übereinstimmung mit Arundinaria auf und so schien es mir berechtigt, unter Berücksichtigung noch anderer Merkmale,—vor allem: Fehlen der in Bastbelege eingeschobenen Parenchymlamellen, die Gestalt der Caryopse, die langkriechenden Rhizome u.s.w.—diese Formengruppe von Bambusa loszutrennen und als eine neue Section in Arundinariew aufzunehmen. Die Aufstellung dieser neuen Formengruppe bietet uns doppeltes Interesse; denn einmal erweist dieselbe, dass die Eintheilung nach der Zahl der Stamen, auf welche man in der Bambuseensystematik ein grosses Gewicht zu legen pflegt, nicht immer durchführbar ist. Andererseits kommt diese Formengruppe²) in ihrer Verbreitung auf Japan³) beschränkt vor.

Wir gehen nun zur Betrachtung des Centralcylinders über. Die Anordnung der Leitbündel weicht, wie es von Ross<sup>4</sup>) nachgewiesen wurde, vom typischen Bau der Monocotylen ab. Zu den normalen peripherischen radialen Bündeln, die ausserordentlich polyarch sind,<sup>5</sup>) kommt noch eine Anzahl der inneren isolierten Hadrom- und Leptomstränge hinzu. Im Querschnitte beliebiger junger Wurzeln bemerkt man innerhalb der dünnwandigen Endodermis ein oder zwei Schichten des ununterbrochenen Pericambiums (Fig. 23), dessen allgemeine Vorkommniss in Bam-

<sup>1)</sup> Derartige Structur findet man nicht in Seh wen dener's Aufzählung der verschiedenen Verstärkungsformen (vergl. *l.e.* p. 132).

<sup>2)</sup> Die Anzahl der bis jetzt bekannten hierhergehörigen Arten ist neun. Vergl. Makino. Bambusaceæ Japonicæ. Bot. Mag. XIV, Nr. 156, p. 20.

<sup>3)</sup> Vielleicht auch in China.

<sup>4)</sup> Ross, Beiträge zur Anatomie abnorm. Monocotylenwurzel. Ber. d. D. B. G. Ed. I, p. 337.

<sup>5)</sup> Z. B. in einer 4 mm dicken Wurzel von *Phyllostachys mitis* habe ich mehr als 150 gezählt.

buseen um so mehr Beachtung verdient, als bei den meisten Gramineen die primordialen Gefässe nach van Tieghem¹) direct der Endodermis anzustossen pflegen. Die inneren Hadromstränge kommen in etwa drei concentrischen Ringen angeordnet vor. Dazwischen liegen zerstreut die inneren Leptomstränge, welche jedesmal aus den 1 oder 2 Siebröhren und den englumigen Geleitzellen bestehen (Fig. 43 etc). Bei echten Bambusa-Arten besitzt die stets einzeln stehende Siebröhre einen regelmässig ovalen Umriss (Fig. 38). Die Gesammtanzahl der inneren Leptomstränge beträgt in den meisten Fällen, wie folgende Beispiele lehren, eine Hälfte der peripherischen, aber bei echten Bambusa-Arten kommen beide fast in gleich grosser Anzahl vor.

		Peripherisehes Leptom	Inneres Leptom
Phyllostachys m	ıitis	84	42
P. $U$	ambusoides	83	41
P. $p$	uberula	47	24
Arundinaria 3	iaponica -	75	40
A.	$\widetilde{Hindsii}$	181	97
A.	Matsumuree	27	13
А. и	ariabilis	30	16
A. $j$	пудтача	43	22
_	palmata	41	20
B.	Veitchii	29	<b>1</b> 5
B. $i$	'amosa	20	10
B. $j$	oaniculata -	39	20
	iipponica	33	16
	vulgaris	70	68
B. $s$	teno <b>st</b> achya	47	41
B. $i$	nana	48	45
Dendrocalamus	s latiflorus	81	55

<sup>1)</sup> Van Tieghem, Les Racine. p. 123; Vergl. Morot, Recherche sur le Pericycle. Ann. d. Sc. nat. Sèr. VI, T. 20, p. 233 und auch Falkenberg, Vergl. Unters. d. Vegetationsorgane. p. 192.

Die übrigen Elemente des Centralcylinders werden, abgesehen vom centralen Markparenchym, prosenchymatisch zugespitzt und zugleich stark verdickt. So entsteht hier ein hohlcylindrischer mechanischer Ring, in welchem sämmtliche Leitstränge eingebettet liegen. Hier muss noch eine Frage gelöst werden: In welcher Weise geschieht die Communication zwischen den einzelnen leitenden Elementen, die von einander getrennt im mechanischen Gewebe liegen? Zwar hat Reinhardt1) die in Frage kommenden Verhältnisse bei den anomal gebauten Wurzeln von Musaceen, Pandanaceen, Palmeen und Cyclanthaceen ermittelt und manch interessantes entdeckt. Betreffs der Communication zwischen einzelnen Leptomsträugen in unserem Fall muss vor allem bemerkt werden, dass die ausserordentlich stark verdickten und verholzten Pericambiumzellen als die Leitungswege zwischen den peripherischen Leptomsträngen kaum in Betracht kommen<sup>2</sup>). Wenn man nun die Zahl der in beliebigen zwei Wurzelquerschnitten vorkommenden Leptomstränge sorgfältig mit einander vergleicht, so kann man leicht eine bedeutende Abnahme derselben nach dem Wurzelspitze wahrnehmen, wie es aus einigen beigefügten Beispielen hervorgeht:

				Zahl der	Zahl der Leptomstränge in				
				Proximalende	Mitte	Distalende			
22.5	em	langes V	Vurzelstück <sup>a</sup>	3) 128	_	104			
12	,,	,,	,,	116	108	102			

Der Umstand beruht bloss darauf, dass die inneren Leptomstränge sich unter einander und mit den peripherischen im weiteren Verlauf allmählig verschmelzen, wie man sich durch Betrachtung successiver Querschnitte überzeugen kann. Die

<sup>1)</sup> Reinhardt, Das leitendegewebe einiger anomalgebauten Monocotylenwurzel. Jahrb. f. wiss. Bot. Bd. XVI, p. 336.

<sup>2)</sup> Reinhardt, l.e. p. 361.

<sup>3)</sup> von Phyllostachys bambusoides.

Figuren 39 und 40 zeigen einige Fälle der erwähnten Verschmelzung. Dieser Modus des Leptomverkehrs ist nach Reinhardt'sehen Angaben auch bei anderen anomal gebauten Wurzeln häufig verwirklicht1). Der zweite Modus ist aber von mehr wirksamer und auffälliger Art. Bei jeder Ansatzstelle der zahlreich entspringenden Nebenwurzeln an Centralcylinder werden sämmtliche hier befindliche peripherische sowie verschieden tief liegende innere Leptomstränge in einem System förmlicher Anastomosenbildung zusammengehalten, welche bei den meisten Arten sieh auf die etwa 10 periperischen Leptomstränge hinüberstreckt und bei Bambusa-Arten sogar die Hälfte des ganzen Umfangs des Centralcylinders in sich umfasst. Die etwas schematisierten Figuren 35 und 36 illustrieren das obengesagte. Das hier die Verbindung zwischen einzelnen Leptomsträngen herstellende Gewebe besteht aus den plasmareichen parenchymatischen Zellen, die mit den ansehnlich grossen Zellkernen und den dünnen unverholzten Wänden versehen sind (Fig. 43 u. 44). Die Verschmelzung zweier Gefässe habe ich nur selten gesehen, während bei der Ansatztelle der Nebenwurzel sämmtliche mechanische Zellen zufolge reichlicher Tüpfelbildung und häufiger auftretender Querwände einen holzparenchymartigen Character annehmen und demgemäss dem Saftaustausch zwischen den eingebetteten Gefässen besser angepasst sind. Den directen Anschluss der Leptomelemente an Holzparenchymzellen, wie es von Reinhardt für Musaceen und Cyclanthaceen<sup>2</sup>) nachgewiesen wurde, habe ich auch häufig bei Bambuswurzeln angetroffen (Fig. 41).

Die Basaltheile des Centralcylinders der Nebenwurzel sind

<sup>1)</sup> Reinhardt, l.c. p. 364, p. 343 etc.

Vergl. Ross, Beitr. z. Anat. abnorm. Monocot. wurzel. p. 334,

<sup>2)</sup> Reinhardt, l.e. p. 343, p. 346 und p. 348.

aus dem stark verdickten porösen rechteckigen parenchymatischen Zellen gebildet, durch welche die kurzen Basalglieder jedes Leptomstrangs abwärts verlaufen, um sich dem oben erwähnten Leptomanastomosencomplex der Hauptwurzel anzuschliessen (Fig. 37). Hingegen scheinen die Gefässe der Nebenwurzel basalwärts meist blind zu endigen, so dass sie nur selten in directen Zusammenhang mit denen der Hauptwurzel kommen.

Die Nebenwurzeln zeigen in ihrem Bau alle Merkmale der bezüglichen Hauptwurzeln, dennoch fehlen ihnen stets die inneren Hadrom- und Leptomstränge (Fig. 47 u. 45).

Die Ansetzung der Wurzeln an die Stammorgane geschiet in der bei Monocotylen üblichen Weise¹). Die Elemente des mechanischen Rings des Wurzelcentraleylinders breiten sich scheibenförmig aus und verschmelzen sich mit den äussersten Bündeln der Stammgebilde. Einzelne losgelöste Wurzelstränge dringen noch weiter ein und schliessen sich den peripheren Stammbündeln an, wobei das Leptom der ersteren solch eine Umgestaltung erfährt, wie sie bei den Knospenbündeln beobachtet wird²).

Es erübrigt noch einen interessanten Befund kurz zu erwähnen. Die Rindenparenchymzellen der Nebenwurzeln, mit Ausnahme von den innersten 2-3 Schichten kleinlumiger Zellen, sind gewöhnlich von einem Pilz bewohnt, der in jedem Zelllumen ein ansehnliches Knäuel von dicken verschlungenen Mycelfäden bildet (Fig. 46 u. 47). Die verpilzten Wurzeln bieten trotzdem ein ganz normales und gesundes Aussehen dar. Die Mycelfäden treiben hie und da sogenannte Vesikulen aus und producieren oft massenhaft gelbe körnige Substanz von nicht genau bekannter

<sup>1)</sup> Vergl. Falkenberg, Vergl. Unters. p. 196.

<sup>2)</sup> Vergl. p. 437.

chemischer Zusammensetzung. Die Stärke verschwindet gewöhnlich vom inficierten Gewebe. Es unterliegt also keinem Zweifel, dass wir in diesem Fall mit einem endotrophischen Mycorhiza<sup>1</sup>) zu thun haben. Der Wurzelpilz fehlte in keiner der von mir untersuchten Arten und ist sowohl in den epidermlosen Nebenwurzeln von Arundinaria- und Phyllostachys-Arten als in den mit Epidermis versehenen Bambusa-Nebenwurzeln constant nachweisbar. Die Rindengewebe der Hauptwurzeln habe ich meist pilzfrei gefunden, abgesehen von den dünneren Wurzeln von Arundinaria variabilis, Bambusa ramosa, etc. Die Lösung der Frage nach der physiologischen Rolle<sup>2</sup>), die dieser Pilzsymbiont in der Ernährung der Baumgräser spielt, will ich mir für künftige Studien vorbehalten.

### DIE BLATTGEBILDE.

Die in zwei entgegengesetzten Reihen gelegenen, breiten Scheideblätter<sup>3</sup>) umhüllen übereinander den ganzen Schössling und auch die wachsende Spitze des Rhizoms.

In der basalen, zum Knotengewebe übergehenden Region jedes Scheideblattes weisen die Leitstränge in ihrem Hadrom kein grosses getüpfeltes Gefäss auf, sondern sie besitzen nur zahlreiche, oft mehr als 15 Ring- oder Spiralgefässe, die mit einander mannigfach anastomosieren. In dem in der mittleren Partie des Scheideblattes ausgeführten Querschnitte erblickt man parallel-

<sup>1)</sup> Frank, Lehrb. d. Botanik, Ed. I, p. 274; Über neue'Mycorhiza-Formen. Ber. d. D. B. G. Ed. V, p. 400.

<sup>2)</sup> Es wurde neuerdings vielfach die Ansicht geäussert, dass die Pflanzen die mit Mycorhiza ausgerüstet sind, der Assimilation des freien Stickstoffs befähigt[seien. Vergl. Janse, Ann. d. Jard. Bot. Buit. Vol. 14, p. 200, und auch Nobbe, Lundw. Versuchs-St. Bd. Ll, p. 241.

<sup>3)</sup> Rivière, Les Bambous, p. 76-82, p. 231.

verlaufende, abwechselnd starke und schwache Leitbündel, die in ihrem Bau kaum von den dem Stammorgan eigenen abweichen (Fig. 48). Sie sind mit einander durch die aus einigen Siebröhren und Gefässen bestehenden Queranastomosen verbunden (Fig. 53). Die Bastbelege auf der Leptomseite stossen gewöhnlich unmittelbar an die stark verdickte Epidermis der Aussenfläche an (Fig. 51), aber bei dicken fleischigen Scheideblättern der Phyllostachys-Arten liegen fast alle Bündel mit ihren Bastbelegen ganz frei im Parenchym (Fig. 52). Entgegengesetzt den stärkeren Bündeln liegen die bandförmigen, meist 2-3 schichtigen Baststränge an der Blattinnenseite. Die letzteren kommen bei Arundinaria Matsumuræ sonst auch an der Blattaussenseite hier und da vor (Fig. 49). Das Scheideblattparenchym besteht aus dünnwandigen, saftreichen Zellen,1) von denen einige subepidermale Schichten bei unterirdischen, harten Scheideblättern sclerenchymatisch verdickt sind. Bei den derben oberirdischen Scheideblättern von Arundinaria-Arten tragen die an die Intercellularräume angrenzenden Flächen der Parenchymzellen die eigenthümlichen bald kugelförmigen, bald stäbehenförmigen Auswüchse, die starke Holzreaction geben (Fig. 54).

Die Spaltöffnungen kommen an der Ober- sowie Unterseite der Scheideblätter vor.

Die laubblatttragenden Blattscheiden stimmen in ihrem Bau mit den oben geschilderten Niederblättern wesentlich überein.

Die Laubblätter der Bambuseen sind schon wiederholt von vielen Forschern anatomisch untersucht worden. So haben Kareltschikoft<sup>2</sup>), Magnus und Haberlandt die Armpallisa-

<sup>1)</sup> Die Parenchymzellen ausgewachsener Scheideblätter enthalten fast keine Stärke, sondern viel Glykose.

<sup>2)</sup> Kareltschikoff, Ub. d. faltenförmig · Verdickungen in d. Zellen einiger Gramineen, p. 180. (Referat).

dennatur des Assimilationsgewebes erkannt. Bei Güntz finden wir Angaben über einige allgemeine Characteristik der Bambuseenblätter, dabei führte das energische Auftreten der mechanischen Elemente ihn zur Aufstellung des "Bambuseentypus" der Gramineenblätter"). Bei dieser Sachlage würde es berechtigt sein, dass ich mich hier nur auf einige kurze Notizen beschränke.

Um jedes Mestombündel bemerkt man zweierlei Scheiden<sup>2</sup>), d.h. eine farblose Parenchymscheide und eine innere verholzte Bastscheide (Fig. 55 u. 56). Die stets einschichtige Parenchymscheide fehlt selbst bei kleinsten Bündeln nicht; bei stärkeren Bündeln ist sie oft dort stark verdickt und verholzt, wo sie an subepidermale Bastrippen anschliesst. Die wenigstens um das Leptom stets vorhandene Bastscheide wurde von Schwendener als Mestomscheide ausgezeichnet<sup>3</sup>) und der echten Schutzscheide zur Seite gestellt. Dieselbe ist um die kleineren Bündel meist einschichtig, aber bei den stärkeren nicht selten mehr als 4 Schichten dick. Ferner verhalten die Elemente dieser Scheide sich gegen Schwefelsäure kaum anders als gewöhnliche verholzte Bastzellen, während sich die unverholzte Parenchymscheide gegen dieses Reagens sehr widerstandsfähig erweist. So ist die in Rede stehende Scheide als eine vereinfachte Form der das Mestom vollkommen umschliessenden Bastscheide, wie ich sie schon bei den Stielbündeln beschrieben habe, aufzufassen.

## IV. Der Entwicklungsvorgang der Schösslinge.

Als Gegenstand der folgenden Darstellung diente mir *Phyllostachys mitis*.

<sup>1)</sup> Güntz, Unters. üb. d. anat. Struct. d. Gramineenbl. p. 64.

<sup>2)</sup> Schwendener, Die Mestomscheiden der Gramineenblätter; Vergl. Strasburger, Leitungsbahnen, p. 344.

<sup>3)</sup> Schwendener, l.c. p. 178.

Die auf jedem Knoten der wachsenden Rhizomspitze angelegte Knospe wird erst im nächsten Jahre zu einem kleinen Schössling mit dem schon differenzierten, verholzten, ca. 1 cm langen Stiel ausgebildet. Diesen letzteren nenne ich kurzweg den Schössling des 2ten Stadiums, während die dem Knoten dicht anliegende, stiellose Knospe als 1stes Stadium von diesem unterschieden wird. Wenn man einen Querschnitt in der oberen Region dieses kleinen Schösslings ansführt, so sieht man den Centralcylinder gesondert in einen peripherischen, schmalen, bündelführenden Ring und in umfangreiches Markgewebe, welches sich nach unten allmählig verschmälert. Auf dem Längsschnitt sieht man dicht unterhalb des Urmeristems vom Vegetationspunkt beginnend eine grosse Anzahl abwechselnd stärkereiche und stärkearme Zonen, welche letztere sich in späteren Stadien zu Internodien verlängern.

Der Schössling des 2ten Stadiums nimmt im Laufe des Sommers an Grösse zu und wächst im Spätherbst (October-November) schon zu einem mittelgrossen Schössling des 3ten Stadiums. In diesem Zustande verharrt er während des Winters.

Anscheinend sehon in März tritt eine rasche Zunahme an Grösse ein und im Anfang April erreichen die Schösslinge unter der Erde eine ansehnliche Grösse, die ich als 4tes Stadium kennzeichnete. Der Schössling ist mit zahlreichen geräumigen, dicken Scheideblättern bedeckt. Der verholzte Stiel ist nun ca. 2 cm lang und 0.9-1.2 cm dick geworden. Die unteren, an den Stiel sich direct anschliessenden, etwa zehn Internodien, deren mittlere Höhe 1-2 cm beträgt, sind mit zahlreichen 3-4 mm dicken und bis etwa 15 cm langen Wurzeln dicht besetzt. Ueber den inneren Bau ist folgendes zu bemerken. Die Spitze, unterhalb des Urmeristems, besteht aus abwechselnd stärkereichen und stärkearmen Zonen, deren Zahl binnen 6 mm 40 beträgt.

Die Dicke der beiden Zonen nimmt nach unten zu, und erst in Entfernung von 1.5-2.0 cm vom Vegetationspunkt entstehen im internodialen Markgewebe sichtbare Querrisse, deren Weite und Höhe in den nachfolgenden Internodien immer zunehmen. Diese primordialen Markhöhlen erreichen in den unteren, mit Wurzeln besetzten Internodien eine maximale Höhe von ca 3 mm, dabei besitzt das Diaphragm eine Dicke von 2.5 mm. Die Bündelanlage in der Spitzenregion besteht aus engen procambialen Zellen. Oberhalb der Stelle, wo die erste Markhöhle zum Vorschein kommt, erfolgt schon die Differenzierung in Elemente des Bündels.

Indessen tritt die Spitze des Schösslings allmählig auf der Erdoberfläche hervor; vom Ende April ab erfolgt dann ein rasches Wachstum desselben. Schon in Mitte Mai erreichen mehrere Schösslinge eine Höhe von S-10 Meter, und an den oberen Nodien findet die Entfaltung von blatttragenden Aesten Mehrere Internodien auf der Erde sind nur 6-9 cm statt. lang und nach oben nimmt die internodiale Länge graduell zu. In mittlerer Höhe der Pflanze erreichen sie die Länge von 20 cm und mehr. Bis auf diese Region haben alle Internodien ihr Längenwachstum vollendet. Mehrere darauf folgende Internodien besitzen basale Wachstumszonen. Die von dort nach oben liegenden Internodien verjüngen sich allmählig zum Vegetationspunkt. Die noch in Streckung begriffenen Internodien sind stets mit Scheideblättern umhüllt. Der Schössling in diesem Zustande ist im 5ten Stadium.

Hier lasse ich einige Zahlenangaben folgen:

		Stadium II.		Sta	Stadium III.			Stadium IV.		
		1	9	3	1	2	3	1	2	3
Länge	Aeussere Curvatur	4.4	4.2	4.6	20.2	19.3	16.0		-	40.5
in Innere cm. Curvatur		3.7	3.8	3.4	13.9	13.4	12.0	56.7	48.0	48.5
Maximal	umfang in em.	5.3	4.9	4.4	15.6	14.8	13.8	34.0	31.5	34.5
Gewie	eht in Gr.	4.18	4.21	4.11	173.0	154.0	116.5	1831.0	1432.0	2062.0

Tägliche Zuwachsmessungen an jedem Mittag vom 24 April

		I.		II.		III.		IV.	
Datu	m.	Länge in cm.	Zuwachs.	Länge in cm.	Zuwachs.	Länge in em.	Zuwachs.	Länge in cm.	Zuwachs.
April	24	34.9		37.2		45.7		27.2	
	25	38.1	3.2	43.1	5.9	51.3	5.6	31.0	3.8
	26	44.2	6.1	50.6	7.5	61.1	9.8	37.6	6.6
	27	53.4	9.2	62.4	11.8	75.0	13.9	46.1	8.5
	28	62.5	9.1	73.3	10.9	91.7	16.7	56.6	10.5
	29	74.1	11.6	87.4	14.1	114.4	22.7	72.9	16.3
	30	86.8	12.7	101.3	13.9 .	135,3	20.9	86.8	13.9
Mai	1	106.9	20.1	123.9	22.6	164.0	28.7	108.8	22.0
	2	137.9	31.0	158.3	34.4	$2\bar{0}5.8$	41.8	141.9	33.1
	3	175.9	38.0	202.4	44.1	253.3	47.5	180.1	38.2
	4	199.1	23,2	230.7	28.3	283.5	30.2	207.7	27.6
	5			277.1	46.4	336.4	52.9	263.2	55.5
	6	300.5	50.7	3÷3.7	56.6	398.6	62.2	319.0	55.8
	7	325.5	25,0	363.7	30.0	418.0	19.4	346.5	27.5
	8	338.6	13.1	375.6	11.9	447.6	29.6	361.2	14.7
	9	382,5	43.9	417.6	42.0	500.6	53.0	409.4	48.2
	10	423.9	41.4	464.5	47.0	546.9	46.3	452.6	43.2
	11	475.3	51.4	515.6	51.1	605.0	58.1	514.1	61.5
	12	551.0	75.7		_		_	597.9	83.8
	13	580.1	29.1	619.2	$51.\overset{*}{8}$	721.4	$58.2^{*}$	636.2	38.3
	14	631.6	51.5	661.9	42.7	786.4	65.0	664.6	28.4
	15	719.6	0.88	744.7	82.8	846.1	59.7	710.7	46.1

NB.-\* Zuwachs ist Mittel von 2 Tagen.

 $<sup>^{\</sup>rm wx}$  Nach Beobachtungen des hiesigen meteorologischen Observatoriums.

Also bei diesen Messungen stieg der maximale Zuwachs nicht selten über 80 cm pro
 Aum. 2. Das bisher bekannte stärkste Wachstum der Eambushalme beträgt 91.3 cm pro
 Aum. 3. Auf die hier beobachteten auffälligen Wachstumsschwankungen und andere interes\_

<sup>1)</sup> Die älteren Angaben über das Wachstum der Bambuspflanzen findet man bei Kraus

Um die erstaunlich grosse Schnelligkeit des Wachstums von Bambusschösslingen in dem 5ten Stadium zu demonstrieren<sup>1</sup>), führe ich hier einige von mir ausgeführte Messungen an *Phyllostachys mitis* an.

von *Phyllostachys*-Halmen, bis 15 Mai ausgeführt.

	V. VI.						
_	Länge in cm.	Zuwachs.	Länge in cm.	Zuwachs.	Wetterangaben.	Mittlere Tempera- tur.**	Mittlere relat. Ilumidi- tät.**
	106.6				klar, leiser Wind	14°.4 C	50.2
	121.2	14.6	52.6		Bar-wenig triib	125.5	73.0
	144.8	28.6	76.3	23.7	klar	15°.4	74.3
	176.8	32.0	108.5	32.2	Hir	16°.4	69.8
	208.3	31.5	140.4	31.9	wenig triib	15°.9	71.3
	250.4	42.1	184.4	44.0	klar, leiser Wind	18°.1	43.5
	284.8	344	216.8	32.4	War, leiser Wind	14°.5	64.9
	324.6	39.8	260.3	43.5	Mar, windig	16°.7	73.0
	378.4	53.8	322.0	61.7	klar, windstill.	18°.4	66.0
	444.4	66.0	288.9	66.9	Regen	17°.6	80.0
	484.2	39.8	429.2	40.3	klar, leiser Wind	13°.8	91.4
	551.7	67.5	493.9	64.7	wen <b>i</b> g triib	17°.5	86.0
	633.9	82.2	566.7	72.8	Regen	17°.7	82.6
	672.4	38.5	607.0	40.3	$Reg \gamma n$	11°.9	93.3
	713.1	40.7	625.4	18.4	halbklar, windig	11°.6	86.6
	755.5	42.4	675.4	50.0	leabhlar, windstill.	15°.6	76.1
	770.5	15.0	738.9	63.5	klar, leiser Wind	16°.6	81.2
					Mar, leiser Wind	17°.8	65.0
					Regen	18°.3	84.8
					klar, leiser Wind.	14°.6	89.6
					1.4ar	18°.5	78.2
					klar	20°.7	69.4

<sup>24</sup> Stunden! Rivière fand denselben bei *Phyllostachys mitis* in Algier 57 cm pro 24 Stunden. Stunden. (Vergl. Pfeffer, Pflanzenphysiologie. Bd. H, p. 83.)

sante Fragen kann ich an dieser Stelle nicht weiter eingehen (Vergl. Kraus, l.c.).

<sup>(</sup>Ann. d. Jard. Bot. de. Buit. vol. XII. p. 197.) zu-ammengestellt.

# V. Verhalten der Baustoffe während der Entwicklung der Schösslinge.

In diesem Kapitel will ich die Umwandlungs- und Wanderungsvorgänge verschiedener Baustoffe während der Entwicklung der Bambusschösslinge in wesentlichen Zügen darzustellen versuchen.

### DIE RESERVESTOFFE.

Unter stickstofffreien Reservestoffen, die sich in Bambuspflanzen vorfinden, kommt die Stärke in erster Linie in Betracht

Im zeitigen Herbst, wo die Ablagerung der Reservestoffe schon stattgefunden hat, konnte ich sie in oberirdischen und unterirdischen Theilen aller untersuchten Arten in wechselnden Mengen auffinden. Die Phyllostachys-Arten, welche mit einem umfangreichen, unterirdischen Rhizomsystem ausgerüstet sind, pflegen nur sehr kleine Mengen der Stärke in ihren Halmparenchymzellen aufzuspeichern. Bei allen Arten wird die grösste Menge der Stärke in Rhizomen und Wurzeln deponiert. Parenchymzellen der Knoten sind stets äusserst stärkereich und die Blattscheiden einiger Arten speichern ebenfalls die Stärke im Parenchym auf. Die Siebröhren sind dagegen höchst inhaltarm; ich habe nur selten winzige Stärkekörner in denselben nach-Es waren aber kleine Mengen von Glykose und gewiesen. Rohrzucker stets vorhanden. Beachtenswerth ist die Gestalt der Stärkekörner; bei Bambusa palmata, B. Veitchii und B. paniculata sind sie aus zahlreichen kleinen Theilkörnern zusammengesetzt (Fig. 57). Solche polyadelphische Stärkekörner<sup>1</sup>) kommen nach Nägeli in Stammgebilden nur selten vor.

<sup>1)</sup> A. Meyer, Untersuchungen über die Stärkekörner, p. 204.

Der reducierende Zucker ist ziemlich reichlich in Halmen und Rhizomen im Winterzustande nachzuweisen. Die winzigen Fetttröpfehen sind oft im Halmparenchym von *Phyllostachys mitis*, Arundinaria Simoni, Arundinaria Hindsii u.s.w. angetroffen, aber sie kommen jedenfalls als Reservestoffe kaum in Betracht.

Um eine Vorstellung über die Mengenverhältnisse der aufgespeicherten Stärke zu anderweitigen Bestandtheilen der Reservestoffbehälter zu gewinnen, habe ich einige Analysen der zweijährigen Rhizome von *Phyllostachys mitis* ausgeführt<sup>1</sup>). Es ergab folgendes:

% Gehalt der
Trockensubstanz.
Stärke24.01
Reducierender Zucker 0.95
Nicht reducierender Zucker 4.31
Rohproteinstoffe $(N \times 6.25)$ 5.41
Fette 0.61
Rohfaser47.32
Asche 8.74
Unbestimmte Stoffe (Differenz) 8.65
100.00

<sup>1)</sup> Das am 25 November gesammelte, kräftige Rhizomstück von Phyllostackys mitis, dessen Parenchym sich zuvor bei mieroscopischer Beobachtung als von Stärke strotzend erwics, wurde mittelst des Hollels abgeschaubt, schnell bei 70°-80° getrocknet, und zu einem feinen Schrot gemahlen. Von diesem lufttrockenen Rhizomschrot wurde ein bestimmtes Quantum abgewogen und zu jeder E stimmung verwendet.

Das Trockengewicht des Schrots wurde nach weiterem 4 stündigen Trocknen bei 100° (zur Gewichtseonstanz) bestimmt.

Die Stärke wurde mittelst der Erhitzung im Soxhlet'schen Antoclave verzuckert.

Die löstlichen Kohlehydrate wurden nach 5-6 maligem Ausziehen mit kaltem Wasser binnen 24 Stunden erschöpft. Der nichtreducierende Zucker wurde nach Inversion mit verdünnter Schwefelsäure bestimmt. Alle Bestimmungen der Zucker wurden nach Meissl-Allihn'scher Gewichtsmethode ausgeführt.

Der Gesammtstickstoffwurde nach Kjeldahl und der Eiweisssticks off nach Stutzer bestimmt.

Die Fasersubstanz wurde durch Weender'sches Verfahren bestimmt.

Das Ätherextract wurde ohne weiteres als Oel angenommen.

Man sieht also, dass die Stärke wohl als Hauptreservestoff zu betrachten ist, dagegen sind die Proteinstoffe in verhältnissmässig geringer Menge vorhanden.

Nun schien es mir erwünscht zu wissen, eine wie weite Strecke des Rhizoms zum Auswachsen eines Schösslings dienen sollte, so habe ich im Anfang Februar von einer Plantation von *Phyllostachys bambusoides* eine Anzahl Rhizomstücke ausgegraben und die auf Knoten vorkommenden Schösslinge (im 3ten Stadium) aufgezählt. Es ergab folgendes Resultat:

Zahl der Rhizomstücke	Gesammtanzahl der Internodien	Halme	Rhizomzweige	Schösslinge
69	632	.)	30	15

Aus obigem berechnete ich das Zahlenverhältniss der Rhizominternodien zu einem Schössling, wie 42.1:1.

### KOHLEHYDRATE.

Die stickstofffreien Reservestoffe in allen untersuchten Arten bestehen, wie sehon erwähnt, hauptsächlich aus der Stärke. Dass der Stärkegehalt der Rhizome eine bemerkbare Verminderung während des Winters erleidet, wie es von Rosenberg¹) für einige perennierende Gewächse dargethan wurde, konnte ich nicht in diesem Fall bestätigen, da ich grosse Anzahl von Rhizomen von Phyllostachys mitis, Phyllostachys bambusoides, Phyllostachys Kumasasa, Arundinaria Hindsii, Arundinaria Narihira, Arundinaria quadrangularis, Arundinaria Matsumuræ, Bambusa palmata, Bambusa nana u.s.w. im Winter (Anfang Januar—Ende Februar)

<sup>1)</sup> Rosenberg, Die Stärke im Winter. Pot. Centralbl. Bd. LXVI, p. 337.

untersuchte und dabei keine merkliche Différenz in Bezug auf Stärkegebalt von den im Herbst beobachteten Exemplaren auffinden konnte. Auch die Wurzeln der untersuchten Arten enthielten in dieser Jahreszeit grosse Mengen von Stärke in ihrem Rinden- und Markparenchym, so z. B. bei den am 25. Januar gesammelten Exemplaren:

	Phyllostachys mitis	Phylles achys bambusoides	Arundinaria Hindsii	- Avundinaria Navihira
Rindenparenchym	$-1^{1}$ )	-1	.5	4
Markparenchym	3	•)	3	3

Gleiches gilt für die oberirdische Halme von Arundinariaund Bambusa-Arten.<sup>2</sup>) Merkwürdigerweise nimmt die Menge des reducierenden Zuckers während des Winters unverkennbar zu. Sodann kann man leicht mehr oder minder bedeutende Mengen desselben im Halm- und Rhizomparenchym obengenannter Arten nachweisen.<sup>3</sup>)

Aber in Stadium IV, wo die unterirdischen Schösslinge ein

<sup>1)</sup> Dequemlichkeitshalber habe ich zur Lezeichnung des Stärkegehaltes folgende Ziffern benatzt:

<sup>0-</sup>bei gänzlicher Abwesenheit von Stärke;

<sup>1-</sup>wenn ein Theil des Gewebes stärkefrei ist, während der andere wenige Körnehen in den Zellen führt;

<sup>2-</sup>wenn alle oder die meisten Zellen wenige Stärkekörner enthalten;

<sup>3-</sup>wenn ein Theil der stärkeführenden Zellen wenige stärkekörner enthält, während der andere recht viel Stärke führt;

<sup>4-</sup>wenn das Gewebe recht viel Stärke enthält:

<sup>5-</sup>wenn alle oder die meisten Zellen strotzend gefüllt sind.

<sup>2)</sup> Vergl. A. Fischer, Beiträge zur Physiologie der Holzgewächse. Jahrb. f. wiss. Eot. Ed. XXII, p. 92, p. 112.

<sup>3)</sup> Dieser Zucker geht aber grösstentheils sehon im Anfang März wieder verloren, ohne dass dabei eine bemerkbare Stärkezunahme stattfand. Auch fielen die Versuche den Zucker in der abgeschnittenen Halmtheilen durch künstliche Erwärmung (im Treibhaus bei 17°–20°C.) zur Stärke überzuführen negativ aus.

rasches Wachstum begannen, ist die deutliche Stärkezunahme in mehreren Rhizominternodien in der Nähe von Knoten, an welchen der wachsende Schössling sitzt, zu beobachten. So z.B. bei *Phyllostachys mitis*:

	Anfang November	Ende December	Anfang Februar	Anfang März	Mitte April
Rindenparenchym	4	4-3	3-4	3-4	5
Centralcylinderparen- chym	-)	2	•)	2	4-5
Markparenchym	:}	7	3-4	3-4	5

Diese Stürkezunahme mag jedoch darauf beruhen, dass die von ferneren Theilen des Rhizoms in Form von Zucker zugeführten Kohlehydrate hier in der Nähe des Schösslings transitorisch in Stärke umgewandelt werden. Dafür sprechen die Umstände, dass erstens in entfernteren Rhizominternodien keine entsprechende Stärkezunahme stattfand, und zweitens schon in dieser Zeit ein Blutungssaft, der eine wichtige Rolle beim Zuckertransport spielt, von jeder beliebigen Schnittfläche des Rhizoms hervorquillt.

Die Stärkezunahme ist vor allem im verholzten Stieltheile des Schösslings ausgeprägt:

	Ende December	Anfang Februar	Anfang März	Mitte April
Rindenparenchym	2-3	2	5-4	5
Centralyclinderparen- chym	3	2	4-3	5
Hadromparenchym	(,)	0	0	0

Gleichzeitig wurde die partielle Entleerung der Rhizomknoten, an welche die Schösslinge sitzen, beobachtet, obgleich die nächst folgenden Internodien, wie schon bemerkt, noch von Stärke erfüllt waren. Von jetzt ab wird die Stärke im Rhizome nach und nach aufgelöst und schon in dem Stadium V, wo die Schösslinge auf der Erde 4-6 Meter hoch wuchsen, verschwinden fast sämmtliche Stärkekörner vom Parenchym der benachbarten Rhizominternodien. So zum Beispiel bei *Phyllostachys mitis*:

## Rhizominternodien-

	16. April	9. Mai	19. Mai
Rindenparenchym	. 2	1	0
Centralcylinderparenchym	5	1-2	O
Markparenchym	ž	4–3	1

## Nodium-

San	16. April	9. Mai	19. Mai
Subepidermale sclerotische Schicht	4	0	0
Rindenparenchym	4	O	О
Centralcylinderparench ym	1-2	О	O

# Stiel des Schösslings-

	16. April	9. Mai	19. Mai
Subepidermale sclerotische Schicht	0	0	0
Rindenparenchym	š	1-0	0
Centralcylinderparenchym	.)	0	0

Hier findet auch in der Wurzel eine entsprechende Stärkeentleerung statt:

	16. April	19. Mai
Rinden- (äusseres grosszelliges	5	0
Rinden- {\text{\ti}\text{\texi{\text{\text{\text{\text{\text{\text{\texi{\texi{\texi{\text{\texi}\tiex{\text{\text{\text{\text{\text{\texi}\tint{\text{\texi}\tint		1-2
Markparenchym	.,-4	O

Merkwürdigerweise konnte ich bei so raschem Auflösen der Stärke eine entsprechende Glykosebildung im Parenchym nicht beobachten; bei der Zuckerprobe nach Schimper erhielt ich nur Spuren von Oxydulkörnern in Parenchymzellen.

Die Kohlehydrate in wachsenden Schösslingen verhalten sich im Grossen und Ganzen analog mit denjenigen in von Sachs, H. de Vries u. A. untersuchten Pflanzen.<sup>1</sup>) Indess ist folgendes noch zu bemerken.

Die Abwesenheit von Stärke im Urmeristem des Vegetationspunktes habe ich im allgemeinen constatiert. Die feinkörnige Stärke wird erst an der Stelle, wo die erste Differenzierung der Bündelanlage und des Grundparenchyms auftritt, nachweisbar und man kann abwechselnd stärkereiche und stärkearme Zonen deutlich sehen. Nach unten tritt der Unterschied im Stärkegehalt dieser abwechselnden Zonen immer schärfer hervor. Der reducierende Zucker tritt an der Spitze weiter unten als Stärke auf und zwar zuerst in dem Marke der internodialen Zone, wo der erste Anfang der Zellstreckung sich durch Zerreissen von Gewebe kund thut. Selbst die fertig gestreckten, unteren Internodien des mehrere Meter hohen Schösslings bleiben noch lange Zeit von der Glykose erfüllt. Sobald die Streckung eines Internodiums vollendet ist, verschwindet die Stärke aus dem Parenchym, abgesehen von einigen winzigen Körnehen in 2-3 Zellen bei Durchlassstellen

<sup>1)</sup> Hier seien nur folgende erwähnt:

Sachs, Physiologische Untersuchung üb. d. Keimung von Schminkbohne.—Sachs, Keimungsgeschichte der Gräser.—De Vries, Wachsaumsgeschichte der Zuckerrübe.— De Vries, Keimungsgeschichte der Kartoffelknollen.—Det mer, Vergl. Physiologie d. Keimungsprocess der Samen.—Hoftmann, Über d. Stoffwanderung bei d. Keimung von Weizen- und Klecsamen.—A. F. A. C. Went, Chemisch-physiologische Untersuchungen üb. d. Zuckerrohr.

<sup>2)</sup> Vergl. Sachs, Üb. d. Stoffe welche d. Material z. Wachstum d. Zellhäute liefern. Jahrb. f. wiss. Bot. Ed. III, p. 207.

der Bündel. Da hierbei sämmtliche Zellwände noch keine nennenswerthe Verdickung zeigen, so erfolgt die weitere Ausbildung der Bündelelemente ohne Gegenwart der umgebenden Stärkescheide, in welcher nach Heine<sup>1</sup>) die nöthigen Baustoffe als Stärke deponiert werden sollen. Die besonders starke Zuckeransammlung in einigen Parenchymschichten um die in Ausbildung begriffenen Bastbelege herum vertritt hier die Stelle der fehlenden Stärkescheide und daher mag sie als Zuckerscheide<sup>2</sup>) bezeichnet werden.

In der wachsenden Wurzel bemerkt man die kleinste Menge der winzigen Stärkekörner nur in der noch zartwandigen Endodermis. Hingegen ist in der Haube von der ersten Anlage die Stärke in ihren Zellen festgehalten.<sup>3</sup>) Der reducierende Zucker kommt in der ganzen Länge der Wurzel, ausser der 4-5 mm langen Strecke der Spitze und der Haube, reichlich vor. Erst in der ca. 40 cm lang gewachsenen Wurzel wird die Abnahme und zuletzt das Versehwinden vom Zucker an der Wurzelbasis bemerkbar.

Wie schon erwähnt konnte ich in Rhizomen und Wurzeln, wo die Reservestärke in Auflösung begriffen war, gewöhnlich nur eine Spur von Glykose auflinden. Analoge Tälle sind bereits bekannt. So z.B. gelangte es Sachs nicht, in Cotyledonen der keimenden *Phascolus*- Samen, in Schildehen von *Triticum* und Zea und auch in Funiculus verschiedener Samen die Glykose nachzuweisen<sup>4</sup>), obgleich hier das Vorhandensein der gelösten

<sup>1)</sup> Heine, Die physiolegische i deutung der sogenannten Stärkescheide. Landw. Versuch-St. 1888, p. 115.

<sup>2</sup> H. de Vries hat früher den Ausdruck im analogen Sinne mit "Leitscheide" Schimper's angewandt.

<sup>3)</sup> Vergl. Sachs, Jahrb. f. wiss. Bot. Bd. III, p. 203.

<sup>4</sup> Sachs, Über die Stoffe, welche das Material zum Wachstum der Zellbäute liefern. Jahrb. f. wiss, Botanik, Bd. 111, p. 248.

Kohlehydrate von vornherein erwartet werden musste. In gewissen Fällen dieser Art ist es nicht unwahrscheinlich, dass die Glykose durch andere lösliche Kohlehydrate ersetzt wird.<sup>1</sup>)

In treibenden Rhizomen von Phyllostachys mitis habe ich nun den Rohrzucker in stärkehaltigen Parenchymzellen mittelst der Invertin-Methode nachgewiesen. Ferner in Rhizomen, von welchen fast alle Stärkekörner schon verschwunden waren (Stadium V), beobachtete ich noch erhebliche Mengen Rohrzucker. Uebrigens habe ich in folgenden Arten den Rohrzucker in Rhizomen während des Austreibens der Schösslinge beobachtet: Phyllostachys bambusoides, Phyllostachys puberula und Arundinaria japonica; und in folgenden im Halmparenchym während des Austreibens der Zweigknospen: Bambusa palmata und Arundinaria japonica. Ferner erhielt ich die Rohrzucker-Reaction in Halmen von Phyllostachys Kumasasa, Arundinaria Simoni, und Arundinaria Hindsii var. graminea. In den Wurzeln von Phyllostachys mitis, deren grosskörnige Reservestärke in Auflösung begriffen war, konnte ich ebenfalls Rohrzucker nachweisen. Gleiches gilt für verholzte Stieltheile der Schösslinge. In allen diesen Fällen kommt Rohrzucker hauptsächlich im Parenchym und viel weiniger in Siebröhren vor. Nun liegt mir der Gedanke nahe, dass in diesem Falle die Kohlehydrate hauptsächlich in Form des Rohrzuckers von Zelle zu Zelle wandern<sup>2</sup>).

Vom Rohrzucker ist noch zu erwähnen, dass ich ihn im

<sup>1)</sup> So z. B. wurde für Gramineen-Scutellum das Vorhandensein vom Rohrzucker anstatt Glykose von Grüss auf micrechemischem Wege sowie auf experimentelle Weise sichergestellt. (Vergl. Ber. d. D. B. G. Bd. XVI, p. 17.). Puriewitsch (Jahrb. f. wiss Bot. Bd. XXXI, p. 53.) hat bei der ersten Periode der Entleerung der Reservestärke das Auftreten nichtreducierenden Zuckers beobachtet. Vergl. Leclare du Sablon, Recherche sur les Reserve Hydrocarbones des Bulbes et des Tuberenles. Rev. gen. d. Bot. 1899.

<sup>2)</sup> Vergl. E. Schulze, Ueber die Verbreitung des Rohrzuckers in den Pflanzen und über seine physiologische Rolle. Zeitschrift f. physiol. Chemie. Bd. XX, p. 552.

jungen Gewebe unterhalb des Urmeristems, wo schon eine Differenzierung in nodiale und internodiale Zonen stattgefunden hat, manchmal, wenn auch nicht immer, durch Invertin-Methode nachgewiesen habe<sup>1</sup>).

### EIWEISS UND AMIDOVERBINDUNGEN.

Im Jahre 1872 hat Pfeffer<sup>2</sup>) zuerst die hohe Bedeutung des Asparagins in der Translocation und Bildung von Eiweiss beim Keimen von Lupinus luteus und einigen anderen Papilionaceen auf microchemischem Wege nachgewiesen. Er hat nämlich constatiert, dass das Asparagin als Auflösungsproduct des Reserveproteins in Cotyledonen entsteht und dann wachsenden Theilen zugeführt wird, und ferner, dass wenn Kohlehydrate bei der Assimilation sich vermehren, das gebildete Asparagin zum Eiweiss regeneriert wird3). Die letzterwähnte Thatsache hat er später durch die Kulturversuche im Dunkeln und in kohlensäurefreier Luft weiter begründet.4) Seit diesen zum ersten Male exact ausgeführten Arbeiten Pfeffer's wurde die Frage nach Eiweissumsetzungen mit immer wachsender Eifrigkeit seitens der Botaniker und Chemiker verfolgt, was zu zahlreichen Arbeiten Veranlassung gab. Schulze und seinen Schülern verdanken wir besonders eine Reihe der Versuche über jene Amidover-

<sup>1)</sup> Ich habe im Gewebe dieser Region eine schöne rosarothe Färbung mit cone. Schwefelsäure erzielt. Diese Reaction deutet darauf hin, dass hier ein lösliches Kohlehydrat neben Eiweis vorhanden ist. Vergl. Frankfurt, Zur Kenntniss der chemischen Zusammensetzung des ruhenden Keimes von Triticum vulgare. Landw. Versuchs-St. 1896. p. 461.

<sup>2)</sup> Pfeffer, Untersuchungen über die Proteinkörner und die Bedeutung des Asparagins. Jahrb. f. wiss. Bot. B4. VIII, p. 429.

<sup>3)</sup> Pfeffer, l.c. p. 558.

<sup>4)</sup> Pfeffer, Über die Beziehung des Lichtes zur Regeneration von Eiweissstoffen aus dem beim Keimungsprocess gebildeten Asparagin. Monatsber. d. Acad. d. Wiss. z. Perlin. Dec. 1873.

bindungen und Hexonbasen, wie Glutamin, Leucin, Tyrosin, Phenylalanin, Arginin u.s.w., die neben Asparagin beim Eiweissumsatz auftreten. Namentlich hat Schulze schon in einer im Jahre 1878 publicierten Arbeit die Ansicht geäussert, dass in Lupinenkeimlingen andere Nichteiweiss-Verbindungen, die neben Asparagin auftreten, auch zur Regeneration des Eiweisses dienen müssen¹). In demselben Jahre hat Borodin eine allgemeine Verbreitung von Asparagin im Pflanzenreiche festgestellt, dabei sprach er aus: "Sobald irgend ein lebenskräftiger Theil irgend einer Pflanze arm an stickstofffreien Substanzen wird, sieht man in ihm Asparagin als Zersetzungsproduct des Eiweisses auftreten und sich mit der Zeit immer mehr anhäufen." Demnächst fand Kellner in jungen Theilen der Gräser eine bedeutende Menge Amide, und äusserte zuerst die Ansicht, dass die Amide durch Synthese aus anorganischen Stickstoffverbindungen entstehen. Auch Hornberger4) meinte, dass die Amide, die in Maiskeimpflanzen auftreten, synthetische Producte seien. Suzuki<sup>5</sup>) hat angegeben, dass er bei Einführung von anorganischen Salzen wie Ammoniumnitrat und Natriumnitrat in verschiedenen Pflanzen eine Asparaginbildung bewerkstelligen konnte. Emmerling<sup>6</sup>) hat auch Amidosäuren als synthetische Producte angesehen, aber es fehlt an einem experimentellen Beweis. So kann die Bildung des Aspara-

<sup>1)</sup> E. Schulze, Über Zersetzung und Neubildung der Eiweissstoffe bei der Keimung von gelber Lupine. (Jahresber, f. Agr. Chem. 1878, p. 211).

<sup>2)</sup> Borodin, Über die physiologische Rolle und die Verbreitung des Asparagins in Pflanzenreich. Bot. Zeit. 1878, p. 826.

<sup>3)</sup> Kellner, Landw. Jahrbücher. Bd. VIII. Suppl. 1879.

<sup>4)</sup> Hornberger, Chemische Untersuchung über das Wachstum der Maispflanze. Landw. Jahrb. 1882.

<sup>5)</sup> Suzuki, On the Formation of Asparagin in Plants under different Conditions. Bull. of the College of Agriculture. Bd. II, p. 409.

<sup>6)</sup> Emmerling; Studien über Eiweissbildung in der Pflanze, Landw. Versuchsst. 1887. p. 7.

gins und der anderen Amidokörper entweder durch Zerfall des Eiweisses oder durch geeignete Synthese erfolgen. Ob diese oder jene geschieht muss von Fall zu Fall bestimmt werden.

Jedenfalls ist es seit Pfeffer's bahnbrechender Untersuchung klar, dass die Amide und Amidosäuren, deren Entstehungen in verschiedenen Fällen verschieden sein können, nachher für Eiweissregeneration verbraucht werden. Gegenwärtig ist es aber noch nicht sicher ob verschiedene Amide und Amidosäuren ganz gleichwerthig für Eiweissregeneration dienen. Zwar hat Hansteen¹) in seiner interessanten Arbeit gezeigt, dass bei Lemna minor verschiedene, künstlich eingeführte Amide und Amidosäuren je nach der Qualität der disponiblen Kohlehydrate sich für Eiweissbildung verschieden verhalten. So liegt der Gedanke nahe, dass die in bestimmten Keimpflanzen auftretenden Amidokörper auch ungleichen Werth für Eiweissbildung besitzen. Früher war Schulze<sup>2</sup>) der Meinung, dass das Asparagin schwerer verwendbar als andere Amidokörper ist und daher in Keimpflanzen zur Anhäufung kommt. Aber Loew<sup>3</sup>) behauptete, dass das Asparagin dem Eiweiss näher steht als andere Amidokörper, und vermuthete auch, dass die letzteren weiter zerfallen unter Bildung von Formaldehyd und Ammoniak, aus denen durch synthetische Processe Asparagin entsteht. Erst neulich ist Schulze4) zu einer ähnlichen Vorstellung gelangt. Er spricht die Ansicht aus, dass das Asparagin (und auch Glutamin) in den

<sup>1)</sup> Hansteen, Beiträge zur Kenntniss der Eiweissbildung und die Bedingung der Realisirung. Ber. d. D. B. G. Bd. XIV, p. 362.

<sup>2)</sup> Schulze, Über den Eiweissumsatz im Pflanzenorganismus. 1880. p. 30.

<sup>3)</sup> O. Loew, The Energy of living Protoplasm. Bulletin of the College of Agriculture Bd. II, p. 64.

<sup>4)</sup> Schulze, Über den Umsatz der Eiweissstoffe in den lebenden Pflanzen. Zeit. f. physiol. Chemie. Bd. XXIV, p. 60.

Schulze, Über die Bildungsweise des Asparagius in den Pflanzen. Landw. Jahrb. 1898. p. 509; p. 513.

Keimpflanzen zum grossen Theil durch Umwandlung der Amidosäuren, die als directe Eiweisszersetzungsproducte betrachtet werden können, entstehen, und dass die Amidosäuren einmal zu leicht verwendbaren Amiden¹) übergeführt werden, bevor sie sich in Eiweiss verwandeln.²) Bei dieser Sachlage ist es wünschenswerth im concreten Falle die Localisation und das Verhalten von Amiden und Amidosäuren zu verfolgen und damit einigermassen Aufschlüsse über die Beziehung zur Eiweissregeneration der beiden verschiedenen Stoffe zu gewinnen.

Bei dem vorliegenden Falle der Entwicklung der Bambusschösslinge kommen Tyrosin und Asparagin reichlich vor. Die genannten Vertreter von beiden Stoffgruppen sind glücklicherweise leicht auf microchemischem Wege bestimmbar.

Hier lasse ich ältere Angaben über das Vorkommen des Tyrosins vorangehen. Gorup-Besanetz<sup>3</sup>) fand es zuerst im Wickenkeimlinge. Schulze und Barbieri<sup>4</sup>) fanden es in etwas grösserer Menge in Kürbiskeimlingen. Auch in Lupinenkeimlingen scheint es nicht zu fehlen, da Belzung<sup>3</sup>) aus Extract der

<sup>1)</sup> Eine entgegengesetzte Meinung, dass das Asparagin ein für Eiweissregeneration wenig geeignetes Material sei, wurde neuerdings wieder von Prianischnikow (Landw. Versuchs-St. 1899. Bd. LH, p. 347 ff.) vertreten.

<sup>2)</sup> Unter neueren Publicationen über Eiweisssynthese, die nach Vollendung meines Manuscriptes in meine Hand gelangten, seien nur folgende zu erwähnen:

Prianischnikow, Eiweiszerfall und Athmung in ihren gegenseitigen Verhältnissen. Landw. Versuchs-St. Bd. LH, p. 137.

Hansteen, Über Eiweisssynthese in grünen Phanerogamen. Jahrb. f. wiss. Bot. Bd. XXXIII, p. 417.

Prianischnikow, Die Rückbildung der Eiweissstoffe aus deren Zerfallspreducten. Landw. Versuchst. 1899. p. 347.

Schulze, Über Eiweisszerfall und Eiweissbildung in der Pflanze, Ber. d. B. G. 1900, Heft. 2. p. 36.

Emmerling, Studien über die Eiweissbildung in der Pflanze. Landw. Versuchs-St. Bd. LIV, p. 215.

<sup>3)</sup> Gorup-Besanetz, Ber. d. D. C. G. VII, p. 146; p. 569.

<sup>4)</sup> Landw. Jahrb. Bd. VII, p. 431.

<sup>5)</sup> Belzung, Recherche sur l. Germination etc. Ann. d. Sc. nat. Bot. Ser. VII, T. 15, p. 234.

Keimlinge von Lupinus luteus Tyrosinkrystalle isolieren konnte, obwohl ihm der microchemische Nachweis des Tyrosins nicht gelang. Ferner fand es Schulze') in Cotyledonen keimender Lupinusarten, etiolierten Keimlingen der Lupinus angustifolius, Endosperm von Ricinus communis und etiolierten Pflanzen von Tropacolum majus. In allen diesen Fällen ist die Menge des gefundenen Tyrosins immer sehr gering, so dass man auf microchemische Verfolgung desselben verzichten muss. bemerkte, dass der Grund des geringen Vorkommens von Tyrosin darin liegt, dass es eine viel regere und schnell verlaufende Umwandlung erleidet.2) In unterirdischen Pflanzentheilen ist Tyrosin öfters auf chemischem Wege gefunden. Schulze und Barbieri fanden Tyrosin neben Leucin in den Kartoffelknollen und in der Wurzel von Beta vulgaris.3) Auch Planta4) fund es in den Knollen von Stachys tuberifera. In der botanischen Litteratur finden wir nur vereinzelte Angaben. Prantl<sup>5</sup>) hat Krystalle, die wie Tyrosin reagierten, aus in Alcohol aufbewahrten Stengeln von Dahlia variabilis erhalten. Borodin<sup>6</sup>) fand in Blättern der etiolierten Kartoffel, die mit absolutem Alcohol behandelt wurden, Tyrosinkrystalle. Ferner fand er dergleichen in Vicia sativa, Tropacolum majus etc. Aber es ist hier zu bemerken, dass diese Befunde ausschliesslich von abgeschnittenen und in Wasser weiter cultivierten Zweigen herrührten und gleichzeitige chemische

<sup>1)</sup> Schulze, Üb. d. Umsatz d. Eiweissstoffe in d. leb. Pflanze. Zeit. f. physiol. Chemie. Bd. XXIV, p. 58.

<sup>2)</sup> Schulze, l.c. p. 50.

<sup>3)</sup> Vergl. Schulze, Über den Eiweissumsatz im Pfanzenorganismus. 1880. p. 24.

<sup>4)</sup> Planta, Über die Zusammensetzung der Knollen von Stachys tuberijera. Landw. Versuchs-St. Bd. 35, p. 473.

Vergl. ferner Schulze, Zeits. f. physiol. Chemie. Ed. XXIV, p. 85.

<sup>5)</sup> Prantl, Das Inulin. 1870. p. 61.

<sup>6)</sup> Borodin, Über die physiologische Rolle und die Verbreitung des Asparagins. Bot. Zeit. 1878. p. 819.

Belege fehlten. Erst später hat er¹) einmal in normalen, jungen Dahlia-Blättern Tyrosin aufgefunden. Noch später hat Leitgeb²) den Gehalt der Dahlia-Knollen an Asparagin und Tyrosin constatiert.

Bevor ich zur Besprechung meiner Beobachtungen fortschreite, will ich hier die Ergebnisse von chemischen Untersuchungen Kozai's") kurz erwähnen. Er hat die Analyse des Schösslings (Stadium IV) von *Phyllostachys mitis* ausgeführt; sie ergab folgendes:

	% Gehalt der
	Trockensubstanz.
Rohproteinstoffe	25.12
Fette	2.49
Rohfaser	11.60
Stärke	3.33
Glykose	8.15
Andere N-freie ext. Stoffe	30.49
Asche	9.22
Unbestimmbare Stoffe	9.60
	100.00

Für die Vertheilung des Stickstoffs auf Proteinstoffe und nichtproteinartige Verbindungen ergaben sich folgende Zahlen:

N in Proteinstoffen	1.22% dei	· Trockensubstanz.
N in nichtproteinartigen Stoffe	en2.82% ,,	"
Gesammtstickstoff	4.04%, ,,	,,

So sieht man, dass die Schösslinge grosse Mengen von stickstoffhaltigen Substanzen enthalten, im auffallenden Gegen-

<sup>1)</sup> Borodin, Über einige bei Bearbeitung von Pflanzenschnitte mit Alcohol entstehende Niederschlag. Bot. Zeit. 1882. p. 589.

<sup>2)</sup> Leitgeb, Der Gehalt der Dahliaknollen an Asparagin und Tyrosin. Mittheil, a. d. bot. Inst. z. Graz. 1888, p. 222.

<sup>3)</sup> Kozai, On the nitrogenous non-albuminous Constituents of Bamboo shoots. Eulletin of the College of Agriculture. Vol. I. No. 7.

satz zum Rhizom, und insbesondere kommen die nichteiweissartigen Verbindungen in überwiegender Quantität vor. Kozai hat nach dem Schulze'schen Quecksilbernitratverfahren die seidenglänzenden Nadelkrystalle aus dem Wasserauszug von Schösslingen erhalten, welche mit Sicherheit mit Tyrosin identificiert wurden. Ferner hat er auch das Asparagin isoliert und durch verschiedene Reactionen und Bestimmung der Stickstoffzahl sicher nachgewiesen.

Bei meinen Studien wurden die obengenannten Substanzen, das Asparagin und das Tyrosin in ihrem Verhalten näher verfolgt. Beide sind nach Borodin'scher Methode reichlich und sicher nachweisbar, dabei scheint der vorhandene Zucker kein Hinderniss zur Krystallisation darzubieten.

Zunächst will ich das Verhalten von Asparagin und Tyrosin bei der Entwicklung der Schösslinge von *Phyllostachys mitis* kurz angeben.

Ich konnte weder Tyrosin noch Asparagin im urmeristematischen Gewebe der ganz jungen Knospen (Stadium I) finden, während in deren basalen, von Bündelanlagen durchsetzten Theilen Tyrosin schon regelmässig vorkommt. Nun die Schösslinge nehmen sehr langsam an Grösse zu und ihre Stieltheile werden, wie schon erwähnt, allmählig verholzt. Ich beobachtete, dass das Tyrosin mit der Zeit im Schösslingskörper erscheint und seine Menge immer grösser wurde, zugleich auch das Asparagin in nachstehender Menge. Wenn man einen 4-5 cm langen Schössling in diesem Stadium (Stadium II) untersucht, so sieht man folgendes: Der Vegetationspunkt bleibt frei von Amidosubstanzen. Erst 2-3 mm unten, wo die Bündelaulagen schon differenziert waren, erscheint die erste Spur von Tyrosin im parenchymatischen Gewebe. Asparagin tritt noch weiter unten ein, wo Zucker

in grösserer Menge vorkommt (etwa in der Mitte von der ganzen Länge des Schösslings), daneben viel Tyrosin. Zuletzt fand ich im Stieltheile keine Amide mehr. So eoineidiert Asparagin in seiner Localization fast mit reducierendem Zucker. Pfeffer¹) bemerkte schon derartiges Zusammentreffen von Traubenzucker und Asparagin in der ersten Periode der Keimung von Lupinus luteus. In diesem und auch im folgenden Stadium konnte ieh weder Tyrosin noch Asparagin im Rhizom nachweisen.

Das oben definierte Stadium III wird im Laufe des Sommers erreicht. Während dieser Zeit nimmt die absolute Menge des Tyrosins sowie des Asparagins immer mehr zu, so dass die Krystalle des Tyrosins und des Asparagins unter dem Microskop in grösserer Menge und viel leichter gefunden werden; die Behandlung der Gewebe (die aber eiweissarm sind) mit Millon's Reagens bringt überall eine tiefere Färbung als in den vorigen Stadien. Die Vertheilung des Tyrosins und des Asparagins stimmt im Wesentlichen mit der des vorigen Stadiums überein. Dabei ist noch zu bemerken, dass das Tyrosin weniger in Nodien als in Internodien vorkommt. In diesem Zustande überwintern die Schösslinge ohne bemerkbare Veränderung bis Ende Februar. Von jetzt ab erwacht ein regerer Process im Schösslinge, und von Anfang-Mitte April wächst es sehon zu einer beträchtlichen Grösse unter der Erde. Die Schösslinge in diesem Stadium (Stadium IV) werden auf dem Markt als Gemüse feil geboten. Die oben angegebene analytische Bestimmung Kozai's rührt auch von einem solchen Schösslinge her. In diesem Stadium bemerkte ich folgende Vertheilung: Der Vegetationspunkt ist frei von Amidokörpern, dagegen reich an Eiweiss, aber in jungen

<sup>1)</sup> Pfeffer, Über die Proteinkörner und die Bedeutung des Asparagins. Jahrb. f. wiss. Bot. Bd. VIII, p. 539.

Scheideblättern, die zu dieser Region gehören, lassen sich stets kleine Mengen Tyrosins nachweisen, daher muss man bei Feststellung der Abwesenheit der Amidosubstanzen in der Spitze sie thunlichst von Scheideblättern befreien. Das Eiweiss, welches Biuretreaction giebt, ist in dieser Region besonders reichlich in Procambialsträngen nachweisbar. Nach Sachs¹) wird das Eiweiss durch diese Gewebe dem Urmeristem zugeführt, und da ich hier in der Spitze keine Amide auffinden konnte, so kann die Wanderung des Eiweisses wohl in dem Sachs'schen Sinne geschehen. Das in dieser Weise von unten zugeführte Eiweiss befindet sich unterhalb des Urmeristems räumlich getrennt von Stärke in regelmässig abwechselnden Zonen von je ca. 0.15 mm Dicke. Diese Zonen deuten schon zukünftige Internodien und Nodien an, und befinden sich in den ersteren Eiweiss und in den letzteren Stärke. Erst 4 mm unter dem Vegetationspunkt tritt die erste Spur von Tyrosin auf und nach unten nimmt es immer in den parenchymatischen Zellen an Menge zu. Zugleich ist die Abnahme des Eiweisses in parenchymatischen Zellen leicht constatierbar. Das Asparagin kommt noch weiter unten (ca. 1-1.5 cm unter dem Vegetationspunkt) fast gleichzeitig mit reducierendem Zucker zum Vorschein. Da dicht unter dieser Region die erste Zerreissung im Markgewebe, die den ersten Anfang der Markhöhle andeutet, stattfindet, so soll hier die Zellstreckung erst recht ausgiebig geworden sein. Nach unten nimmt die Menge des Tyrosins und des Asparagins stetig zu, und dabei übertrifft die Menge des Tyrosins bedeutend die des Asparagins. Am reichlichsten findet man

<sup>1)</sup> Sachs, Über die Leitung der plastischen Stoffe durch verschiedene Gewebeformen. Flora, 1863.

Es ist bekannt, dass das Eiweiss unter Umständen die Cellulosemembran hindurch diosmiren kann. Vergl. Puriewitsch, Physiol. Unters. üb. Entleerung der Reservestoffbehälter. Jahrb. f. wiss. Bot. XXXI, p. 68; Pfeffer, Pflanzenphysiologie. Bd. I, p. 613.

Tyrosin an Stellen, wo die Wurzelanlagen zur Bildung kommen,¹) so dass Tyrosin beim Schneiden des Gewebes mit dem Messer sofort im Zelleinneren zu Krystallen erstarrt.²) In den Bastzellanlagen der Gefässbündel, welche noch keine Wandverdickung zeigen, kommt das Tyrosin bedeutend reichlicher als im Parenchym vor. Die Ueberreste des Markparenchyms enthalten nur sehr wenig Tyrosin. Das Millon's Reagens bewirkt stark blutrothe Färbung des Zellsaftes, entsprechend dem hohen Gehalt an Tyrosin. Jedenfalls hat die absolute Menge des Tyrosins im Vergleich mit den vorigen Stadien bedeutend zugenommen. In dieser Region dagegen konnte ich das Asparagin nur mit Schwierigkeit auffinden. Es sei noch hervorzuheben, dass das Asparagin in der Regel in Knoten und Diaphragmen sich nicht befindet, dagegen fehlt es hier an Tyrosin nicht.

In den untersten Internodien, wo die Verholzung der Bastelemente schon eingetreten ist, verliert sich auch das Tyrosin.

Im Laufe des Aprils durchbrechen die Schösslinge einer nach dem andern die Erde und wachsen ungemein rasch in die Länge. Es ist nicht zu bewundern, dass in so schnell wachsenden Pflanzentheilen ein ausgiebiger Eiweissumsatz vor sieh geht. Tyrosin und Asparagin sind sehr reichlich in den oberen wachsenden Internodien vorhanden, mit gleicher Vertheilungsweise wie im vorigen Stadium, d.h. Tyrosin tritt ca. 2 em unter dem Vegetationspunkt auf und Asparagin ca. 4 cm unter demselben gleichzeitig mit reducierendem Zucker. Aber sehr interessant ist die Vertheilungsweise in halberwachsenen Internodien. Nämlich in der unteren weichen Wachsthumszone eines solchen Internodiums befindet sich das Asparagin ziemlich viel neben reichlichem

<sup>1)</sup> ca. 10te Internodium von unten-

<sup>2)</sup> Siehe unten p. 482.

Tyrosin. Hingegen der obere, schon erwachsene Theil desselben Internodiums enthält kein Asparagin, aber Tyrosin in einer nahezu gleich grossen Menge wie im unteren weichen Gewebe. Hier werden Tyrosinkrystalle in jungen Bastzellen, die eben die erste Verdickung begonnen haben, reichlich ausgeschieden, aber es befindet sich viel weniger in Parenchymzellen. Ferner enthalten die Hadrom- und Leptomelemente niemals Tyrosin. Die Nodien und Diaphragmen enthalten weniger Tvrosin als in Internodien und gewöhnlich kein Asparagin. In den eben im Wachstum vollendeten Internodien kommt Tyrosin noch in fast gleich grosser Menge vor, aber Asparagin nicht mehr. In weiter unten liegenden älteren Internodien und Nodien verschwindet allmählig auch das Tyrosin, und mehrere ganz erwachsene und in Verholzung begriffene Internodien auf der Erdoberfläche sind durchgehends von Amidokörpern frei, obwohl sie noch reichlich die Glykose, wie schon bemerkt, im Parenchym aufspeichern.

Von den Scheideblättern habe ich hier nur zu erwähnen, dass sich in der basalen, weichen Wachstumszone jedes Blattes ziemlich viel Asparagin neben reichlichem Tyrosin befindet, und das erstere verliert sich schon an der Uebergangsstelle zu harten Theilen, während Tyrosin noch weiter oben im Parenchym der erwachsenen Spreitentheile reichlich vorkommt. So bemerkt man hier ein ganz ähnliches Verhältniss wie im Halm.

In einige mm hoher Wurzelanlage, sowohl in Periblem wie in Plerom, lässt sich fast kein Tyrosin nachweisen, obwohl dicht darunter liegendes Knotengewebe an demselben reich ist. In verschieden langen wachsenden Wurzeln ist die Spitze stets tyrosinfrei, und erst 1.5-2 cm unten ist eine Spur nachweisbar. Allerdings kommt Tyrosin nur in sehr kleiner Menge im Wurzelparenchym vor, so dass die microchemische Nachweisung

immer schwierig ausführbar ist. Noch spärlicher kommt Asparagin in wachsender Region vor. Diese Umstände können zum Theil dadurch erklärt werden, dass die Wurzeln nur langsam wachsen und demgemäss hier der ausgiebige Eiweissumsatz nicht stattfindet.

Die mit dem oben angegebenen ganz übereinstimmende Vertheilungsweise des Asparagins und des Tyrosins habe ich auch in Schösslingen folgender Arten constatiert:

> Phyllostachys bambusoides, Phyllostachys puberula, Bambusa palmata.

In den von mir in dieser Beziehung untersuchten Arundinaria-Arten, nämlich:

Arundinaria japonica, Arundinaria quadrangularis, Arundinaria Matsumura, Arundinaria Hindsii,

zeigte Asparagin auch das gleiche Verhalten, während ich Tyrosin nur schwierig auffinden konnte, in auffallendem Gegensatz zu *Phyllostachys*-Arten. Wie dies zu Stande kommt ist mir unbekannt.

Ferner ist hier zu bemerken, dass ich in Rhizomen von Bambusa palmata Asparagin in geringer Menge nachweisen konnte, während es mir bei Phyllostachys-Arten nicht gelang.

Aus dem oben erörterten Befunde lasse ich folgende vier Sätze gelten, nämlich:

- 1. Tyrosin übertrifft Asparagin in Menge.
- 2. Tyrosin tritt in der Nähe vom Vegetationspunkt auf, dagegen kommt Asparagin noch weiter unten gleichzeitig mit Glykose zum Vorschein.

- 3. Asparagin verschwindet aus Nodien und Internodien sobald ihre Streckung aufhört, während Tyrosin noch lange Zeit in denselben erhalten bleibt.
- 4. Tyrosin verschwindet zuletzt aus ganz erwachsenen und in Zellwandverdickung begriffenen Internodien und Nodien.

Nun müssen die einmal vorhandenen und allmählig verschwindenden Amidokörper, wie sehon bekannt, zur Eiweissregeneration, sei es direct oder indirect, verwendet werden, weil sonst weitere Zersetzungs- oder Oxydationsproducte, wie Ammoniak, Nitrate u.s.w. in den Schösslingsgeweben angehäuft werden müssen, was durchaus nicht der Fall ist. Zwar habe ich weder Ammoniak noch Nitrat mit Nessler's Reagens resp. Diphenylamin-Schwefelsäure in den betreffenden Geweben nachgewiesen.

Bei der Betheiligung an diesem Eiweissregenerationsprocess scheint, wie aus den oben erwähnten Thatsachen ersichtlich ist, Asparagin viel leichter verwendbar zu sein und so kommt es nur an Stellen, wo regere Eiweissbildungsprocesse stattfinden, vor. Auch in diesem Sinne lassen sich die folgende Beobachtungen erklären. In verkümmerten Schösslingen von Phyllostachys mitis, die täglich nur einige mm wachsen, während nebenbei stehende kräftige Exemplare täglichen Zuwachs von mehr als 70 cm zeigten, konnte ich in verschiedenen Internodien Asparagin niemals auffinden, dagegen kam Tyrosin dort reichlich vor. Ganz ähnlich verhalten sich die Rhizomspitzen von Phyllostachys mitis und Phyllostachys bambusoides, die im Spätherbst (October) untersucht wurden, wobei sie äusserst langsam wachsen. In verschiedenen Internodien derselben konnte ich trotz vielfacher Bemühungen kein Asparagin mit Sicherheit nachweisen, während Tyrosin dort reichlich vorkommt. Dass das Vorkommen von

Asparagin stets mit der lebhaften Stoffbildung bei schneller Streckung verbunden ist, ergiebt sich auch aus folgender Bemerkung Pfeffers¹): "War früher die Wurzel das lebhaftest wachsende Organ des Keimpflünzehens, so ist dieses jetzt das Stämmehen geworden und dementsprechend wendet sich jetzt der Hauptstrom von Glykose und Asparagin in dieses."

Hingegen verhält sich das Tyrosin viel träger in dieser Beziehung, so dass es in schon erwachsenen Theilen lange Zeit züruckbleibt. Beim Verschwinden des Tyrosins aus ganz erwachsenem Internodium wird ein Theil in loco verbraucht, aber ein anderer Theil wird vielleicht den oberen wachsenden Internodien zugeführt und dabei müssen die jungen Bastelemente als Leitungsbahnen benutzt werden, wie besonders reichlicher Gehalt an Tyrosin es vermuthen lässt.

In jeder Hinsicht sind Asparagin und Tyrosin nicht von gleichem Werthe. Das erste ist ausgezeichneter Eiweissbaustoff. während das zweite es nur bis zu einem gewissen Grade ist, Soweit es leichte Verwendbarkeit des Asparagins anbetrifft, steht mein Ergebniss mit Hansteen<sup>1</sup>) im Einklang.

Wie entstehen das Asparagin und das Tyrosin?

Aus der Localisation ergiebt es sich schon, dass das Tyrosin nur bei der Zersetzung des schon vorhandenen Eiweisses entsteht und nicht durch synthetischen Process. In den jungen Geweben unterhalb des Urmeristems, wo noch kein reducierender Zucker vorhanden ist, tritt es schon auf und vermehrt sich nach unten, in dem Masse wie das Eiweiss in den Zellen abnimmt. Andererseits konnte ich in fungierenden Wurzeln und Rhizomen,

<sup>1)</sup> Pfe ffer, Untersuchungen über die Proteinkörner und die Bedeutung des Asparagins. Jahrb. f. wiss. Bot. Bd. VIII, p. 548.

<sup>2)</sup> Hansteen, Über Eiweisssynthese in grünen Phanerogamen. Jahrb. f. wiss. Bot. Bd. XXXIII, p. 449, p. 485.

die wohl als Bildungsstätte der organischen Stiekstoffverbindungen betrachtet werden dürfen, in allen untersuchten Fällen niemals Tyrosin nachweisen. Ferner giebt der Blutungssaft, der beim Stoffabfuhr vom Rhizome eine wichtige Rolle spielt, keine Tyrosinreaction. Dafür sprechen noch folgende Versuche, die ich wiederholt ausgeführt habe. Wenn man irgend eine abgeschnittene Rhizomspitze oder einen Schössling von Phyllostachys mitis, Phyllostachys bambusoides oder Phyllostachys puberula in destilliertes Wasser stellt und am Lieht oder im Dunkeln verweilen lässt, so sieht man nach 2-5 Tagen bedeutende Zunahme von Tyrosin in beliebigen Internodien. Da in diesem Falle vorher weder Nitrat noch Ammoniak in den Zellen nachweisbar war, so ist die nachträgliche synthetische Bildung von Tyrosin wohl ausgesehlossen, und die beobachtete Tyrosinzunahme muss allerdings auf die Eiweisszersetzung zurückgeführt werden.

Mit dem Asparagin ist die Sache schwieriger zu entscheiden. Obwohl ich in oben erwähnten Versuchen die gleichzeitige Asparaginbildung gewöhnlich nicht beobachten konnte, ist es natürlich nicht ausgeschlossen, dass bei der Eiweisszersetzung hierbei entstandenes Asparagin schnell zur Eiweissregeneration verbraucht wurde und demgemäss nicht in gleichem Masse wie Tyrosin zur Anhäufung kam. Andererseits mag die oben erwähnte Localisation des Asparagins in kräftig wachsenden Theilen dadurch zu Stande gekommen sein, dass das im Rhizome fortwährend gebildete Asparagin mit dem Blutungssaft den wachsenden Schösslingen zugeführt und in den betreffenden Theilen angehäuft wurde<sup>1</sup>). Gegenwärtig haben wir drei Möglichkeiten in Bezug auf Asparaginbildung: erstens durch directe Eiweiss-

<sup>1)</sup> Natürlich muss das Material der in wachsenden Schösslingen auf eine so erhebliche Weise umgesetzten Eiweissstoffe von Rhizomen entstammen.

zersetzung,<sup>1</sup>) zweitens durch Synthese aus Ammoniak<sup>2</sup>) und drittens durch Umwandlung von Amidosäuren etc.<sup>3</sup>) Ob die eine oder andere von diesen Möglichkeiten in unserem Falle zutrifft muss vorläufig unentschieden bleiben.

Nun gehe ich zur Besprechung der interessanten Löslichkeitsverhältnisse des Tyrosins über. Bei der Untersuchung der Schösslinge von Phyllostachys mitis im IV und V Stadium habe ich gefunden, dass alle jungen, noch mit plasmatischem Wandbeleg versehenen Bastelemente und oft auch parenchymatische Zellen mit schönen Tyrosin-Nadelbüscheln erfüllt sind (Fig. 59 u. 60). Dieser Umstand liess mich zuerst vermuthen, dass das Tyrosin schon in den lebenden Zellen in Krystallform vorkommt. Aber nach genaueren Untersuchungen lässt es sich bald feststellen, dass das Tyrosin in den intacten lebenden Zellen ganz gelöst im Zellsaft vorkommt und nur erst in den beim Schneiden geöffneten Zellen zu Krystallen erstarit. Man kann diese Thatsache mit aller Bestimmtheit in folgender Weise beweisen: ein 3-4-zelllagendicker Längsschnitt des tyrosinhaltigen Internodialgewebes wird zuerst durch Herumschwenken in Wasser von den an den Schnittflächen anhaftenden Tyrosinkrystallen befreit und dann unter dem Microskop mit feiner Nadel zerzupft, so sieht man bald, dass in vorher klarem Zellsaft der verletzten Bastelemente und Parenchymzellen eine Krystallbildung stattfindet, welche nach wenigen Secunden sich als Tyrosin deutlich erkennen lässt. wird hier das äusserst schwerlösliche Tyrosin<sup>4</sup>) in so hohem Masse im Zellsaft in Lösung gehalten, dass es sich schon nach

<sup>1)</sup> Pfeffer, Pflanzenphysiologie. Ed. I, p. 464.

<sup>2)</sup> O. Loew, Die chemische Energie der lebenden Zelle. p. 77; p. 78.

<sup>3)</sup> E. Schulze, Üb. d. Umsatz d. Eiweissstoffe in der lebenden Pflanzen. Zeit. f. physiol. Chemie. Bd. XXIV, p. 63.

<sup>4) 1</sup> Theil Tyrosin ist löslich in 1900 Theil Wasser bei 16°C.

blosser mechanischer Verletzung der Protoplasten als Krystalle abscheiden lässt. Die Tödtung des Gewebes durch Chloroformdampf, Osmiumsäuredampf sowie Erhitzung bewirkt ebenfalls die Abscheidung der Tyrosinkrystalle. Ferner kommt in den durch 3% KNO<sub>3</sub>-Lösung sehr stark plasmolysierten Bastzellen Tyrosin nach langer Zeit nicht als Krystalle zum Vorschein¹), aber nach Verletzung der plasmolysierten Zellen treten die Krystalle bald Wie solch eine hohe Löslichheit des Tyrosins zu Stande kommt ist schwer zu beantworten<sup>2</sup>). Allerdings muss hier der Einfluss von den Saftraum umgebenden, lebenden Protoplasten in erster Linie in Betracht kommen. Die Ausscheidung von Tyrosinkrystallen beim Schneiden des Gewebes lässt sich nicht in Schösslingen von I, II und III Stadien zeigen. Ebenso verhalten sich die tyrosinarmen Schösslinge von Arundinaria- und Bambusa-Die Schösslinge von Phyllostachys puberula und Phyllostachys bambusoides zeigen ganz gleiches Verhalten wie im oben dargestellten Fall von Phyllostachys mitis.

Ausserdem scheint sich ein Theil des Tyrosins auch in die Zellwände einzulagern, da die Zellwände der jugendlichen Bastelemente und später auch des Parenchyms immer stärker roth durch Millon's Reagens gefärbt werden, in dem Masse, dass Tyrosin in den Zellen selbst abnimmt. Bekanntlich haben früher Correns³) und Fischer⁴) die Vermuthung ausgesprochen, dass die sogenannte Eiweissreaction der Zellwände verschiedener Pflanzen von in dieselben eingelagertem Tyrosin herrühre. Neuer-

<sup>1)</sup> Vergl. Pfeffer, Pflanzenphysiologie Ed. I, p. 465.

<sup>2)</sup> Jedenfalls ist die Ansicht Belzung's (Recherche chimique sur 1. Germination etc. p. 219), dass das im Zellsaft gelöste Eiweiss die Krystallisation von Asparagin, Leucin etc verhindern soll, ganz unzutreffend, denn in unserem Falle wird die Krystallisation schon durch blosse mechanische Verletzung des Protoplastes im Zellsaft eingeleitet.

<sup>3)</sup> Correns, Über die vegetabilische Zellmembran. Jahrb. f. wiss. Bot. Ed. XXVI, p. 616.

<sup>4)</sup> Fischer, Zur Eiweissreaction der Zellmembran. Ber. d. D. Bot. Gesells. Bd. V, p. 429.

dings vertritt aber Czapek¹) die Ansicht, dass es sich um die Reaction eines phenolartigen Körpers handelt, welchen er dem von ihm in Mooszellwänden aufgefundenen Sphagnol als nahe verwandt betrachtet.

## GERBSTOFFE UND FETTE.

Gerbstoffe und Fette spielen bei Entwicklung der Bambusschösslinge nur eine untergeordnete Rolle.

Bei den von mir untersuchten Phyllostachys- und Bambusa-Arten sind Gerbstoffe in verschiedenen Theilen der Schösslinge überhaupt nicht in nachweisbarer Menge vorhanden.<sup>2</sup>) In dieser Hinsicht bietet Arundinaria quadrangularis ein abweichendes Verhalten dar. Ein ca. 100 em langer Schössling zeigte folgende Vertheilung der Gerbstoffe: Das Urmeristem des Vegetationspunktes ist frei davon, und dann treten sie plötzlich in reichlicher Menge im Niveau der 3ten Scheideblattanlage auf, zugleich mit der ersten nachweisbaren Stärke. Gerbstoffe kommen in einigen nachfolgenden Internodien in gleich reichlicher Menge yor und dann nehmen sie nach unten ab. Dabei reagieren am stärksten die Rindenzellen und die peripherischen Centralcylinderparenchymzellen. Selbst in erwachsenen Internodien und Nodien in der Nähe der Erdoberfläche ist eine schwache Reaction be-Die Wurzelhaube enthält auch kleine Mengen der eisenbläuenden Gerbstoffe. In der dritten oder vierten Scheideblattanlage am Vegetationspunkt treten die Gerbstoffe auf und nehmen nach unten zu. So sieht man hier eine analoge Vertheilungsweise wie in den bisher bekannten Fällen, z. B. bei Vicia,

<sup>1)</sup> Czapek, Zur Chemie der Zellmembran bei den Laub- und Lebermoosen. Flora. 1899-Bd. 86, H. 4.

<sup>2</sup> Abgesehen von kleinen Mengen in Wurzelhauben, Scheideblättern etc.

Helianthus<sup>1</sup>), Zuckerrohr<sup>2</sup>) u.s.w. Im vorliegenden Falle seheinen die autochthonen<sup>3</sup>) Gerbstoffe sich aplastisch zu verhalten, weil hier Zucker, Stärke u.a. auf ganz gleiche Weise vorkommen wie bei anderen gerbstofffreien Arten.

Auch kommt den Fetten höchstens nur eine locale Bedeutung zu als Baumaterial der verkorkenden oder verholzenden Zellwände, so zum Beispiel verschwinden die kleinen Fetttropfen in einer subepidermalen Zellschicht der Wurzel schon bei eintretender Verkorkung der Zellwände. In jeder älteren Halmparenchymzelle von Arundinaria japonica, A. Hindsii, Bambusa floribunda etc. befindet sich je ein ölartiger gelber Tropfen, meist in Verbindung mit Calciumoxalatdrusen. Diese kugeligen Gebilde unterscheiden sich von echten Fetttropfen dadurch, dass sie niemals mit Alkannatinktur sich färben. In vielen Punkten stimmen sie mit dem zuerst von Monteverde<sup>4</sup>) in Gramineen aufgefundenen "Harzkörper" überein.

## Mineralstoffe.

Ich habe in verschiedenen Jahreszeiten die Vertheilung der Mineralstoffe in den Reservestoffbehältern und den Schösslingen verfolgt. Als Untersuchungsmaterial dienten mir hauptsächlich Phyllostachys mitis und Phyllostachys bambusoides.

Ich konnte in den Rhizomen, die schon beträchtliche Quantität der Stärke aufgespeichert haben, die Mineralstoffe leicht auffinden. Sie zeigten folgende Vertheilung sowohl in Internodien als in Nodien:

<sup>1)</sup> Vergl. Kutscher, Über die Verwendung der Gerbsäure im Stoffwechsel der Pflanzen. Flora. 1883, p. 33.

<sup>2)</sup> Went, Chemisch-physiologische Untersuchungen über das Zuckerrohr-Jahrb. f. wiss. Bot. Bd. XXXI, p. 297.

<sup>3)</sup> Kraus, Grundlinien zu einer Physiologie des Gerbstoffes. p. 58.

<sup>4)</sup> Monteverde, Über Ablagerung von Calcium- und Magnesiumoxalat in der Pflanze. Bot. Centralb. 1890. Bd. XLIII, p. 327.

Phosphor reichlich im Parenchym des Centralcylinders; nur wenig in Siebröhren.

Magnesium reichlich vorzugsweise in Siebröhren.

Kalium reichlich im Parenchym.

Calcium nicht nachweisbar in Schnitten; nur kleine Mengen in Aschen.

Schwefel nur spurweise in Schnitten.1)

Chlor ziemlich reichlich im Parenchym; aber fast keins in Siebröhren und anderen Elementen der Gefässbündel.

In den Wurzeln können die obengenannten Stoffe auf übereinstimmende Weise aufgefunden werden. Die Nitrate<sup>2</sup>) sind in Rhizomen nur sehr wenig vorhanden, aber zeitweilig etwas mehr in der Wurzelrinde. Sie kommen hier also überhaupt nicht zur nennenswerthen Aufspeicherung. Leitgeb<sup>3</sup>) hat früher gezeigt, dass der Phosphor in Dahliaknollen hauptsächlich als Calciumphosphat vorkommt. Hier ist aber dies nicht der Fall, da ich keine lösliche Ca-Verbindung in den Zellen auffinden konnte. Die Siebröhren der Rhizome und Wurzeln enthalten, wie schon erwähnt, nur geringe Mengen des Phosphors, dagegen reichlich Magnesia. Daher scheint Magnesium theils als Phosphat, theils als lockere organische Verbindung in Siebröhren vorzukommen. Schimper<sup>3</sup>) und Zacharias<sup>5</sup>) haben auch im Siebröhrensafte von Cucurbita, Wistaria, Aristolochia und Menispermum reichliche Mengen des Magnesiums aufgefunden.

<sup>1)</sup> Schimper (Zur Frage der Assimilation der Mineralsalze, Flora, 1890, p. 223) hat auch in den meisten Rhizomen keine Sulfatreaction erhalten und es dem Vorhandensein von Krystallisation verhindernden Substanzen in Zellen zugeschrieben.

<sup>2)</sup> Vergl. Molisch, Über mierochem. Nachweis von Nitraten. Ber. d. D. Bot. G. Bd. I, p. 154.

<sup>3)</sup> Leitgeb, Über die durch Alcohol in *Dahlia*-Knollen hervorgerufenen Krystalle. Bot. Zeit. 1887. p. 29.

<sup>4)</sup> Schimper, Zur Frage der Assimilation der Mineralsalze. Flora, 1890, p. 228.

<sup>5)</sup> Zacharias, Über d. Inhalt d. Siebröhren von Cueurbita. Bot. Zeit. 1884. p. 71.

Sehritt für Schritt mit der Entleerung der Kohlehydrate versehwinden auch die Mineralstoffe aus dem Rhizomparenehym.

Die Nitrate sind schon im IV Stadium in Rhizomen und Wurzeln nicht mehr nachweisbar. Ebensowenig konnte ich im Blutungssaft die Nitratreaction erhalten. Wahrscheinlich werden die Nitrate hierbei fortwährend zu organischen Verbindungen verarbeitet.<sup>1</sup>)

An den unteren, mit zahlreichen jungen Wurzeln besetzten Theilen der Schösslinge besitzen Phosphor und Kalium eine andere Vertheilungsweise als im Rhizome; sie kommen nun reichlicher in den Bündeln vor. Das Magnesium befindet sich hier auch in Siebröhren bevorzugt. Ich konnte niemals die Nitrate im Schösslingskörper auffinden.<sup>2</sup>) Da sie auch im Rhizome fehlen, so ist es höchst wahrscheinlich, dass überhaupt nur wenig Nitrat als solches in den Schössling eingeführt wird.

Von dieser Region nimmt die direct nachweisbare Menge von Phosphor, Magnesium, Kalium und Schwefel oben nach dem Vegetationspunkte hin immer mehr zu, wie man sich durch die Musterung successiver Querschnitte überzeugen kann. Phosphor kommt anscheinend am reichlichsten in 2-3 em Entfernung vom Vegetationspunkt vor, und von hier nach oben nimmt die direct nachweisbare Menge desselben wieder ab. Merkwürdigerweise kommt Phosphor fast ausschliesslich in den eiweissreichen Procambialsträngen vor.<sup>3</sup>) Im Urmeristem lassen sich die anorgani-

<sup>1)</sup> Die feinen Nebenwurzeln geben stets mehr oder minder starke Nitratreaction. Ob sich in irgend einer Weise der hier befindliche Pilzsymbiont an der Stickstoffassimilation betheiligt, muss zur Zeit dahingestellt bleiben.

<sup>2)</sup> Übrigens ist es klar, dass die Nitrate sich nicht bei so lebhafter Eiweisszersetzung bilden. (Vergl. Schulze, Über d. Vorkommen von Nitraten in Keimpflanzen. Zeit. f. physiol. Chemie. Bd. XXII, p. 83).

<sup>3)</sup> Ich habe auch die Lilienfeld'sche Methode für Erkennung der Localisation des Phosphors mit Erfolg benutzt. (vergl. Strasburger, Das botanische Practicum. III. Aufl. p. 144).

schen Phosphorverbindungen nicht mehr nachweisen, sondern grosse Mengen organischer Verbindungen<sup>1</sup>). Magnesium tritt ebenfalls in der Nähe der Spitze fast ausschliesslich in den Bündelanlagen auf, aber kleine Mengen sind auch in den eiweisshaltigen Internodialzonen vorhanden. Beachtet man die bevorzugten Vorkommnisse des Magnesiums in Siebröhren, Bündelanlagen, Internodialzonen und auch im Urmeristem, so darf man wohl annehmen, dass es irgend eine wichtige Rolle bei Eiweissumsatz oder Eiweisswanderung spielt.2) Kalium lässt sich in der Asche des Vegetationspunktes reichlich nachweisen. Hingegen ist die Schwefelsäure nicht direct im Vegetationspunkt nachweisbar, obgleich sich sehon ca. 2 em weiter unten eine ziemlich starke Reaction zeigt. Das in minimaler Menge zugeführte Calcium wird in einiger Entfernung vom Vegetationspunkte als Kalkoxalat niedergeschlagen. Chlor lässt sich ziemlich viel in eiweissreichen Internodialzonen nachweisen. Im Urmeristem scheint es jedoch gänzlich zu fehlen.

Wenn die Schösslinge über die Erde emporwachsen und die Internodien nach einander ihre definitive Länge erreichen, so nimmt die nachweisbare Menge der Mineralstoffe in denselben stetig ab. In unterirdischen Internodien tritt nun mehr oder minder starke Nitratreaction ein, da die erwachsenen Wurzeln an dieser Region schon ihre Thätigkeit entfalteten und begannen die Bodensalze aufzunehmen.

In der ersten Entwicklungsphase der Wurzel ist eine starke Ansammlung der Mineralstoffe im meristematischen Gewebe leicht constatierbar. In etwas länger erstreckten Wurzeln findet eine ähnliche Ansammlung in der Spitze statt. Phosphor und Mag-

<sup>1)</sup> Vergl. Schimper, l.c. p. 224.

<sup>2)</sup> Vergl. Hornberger, Chemische Untersuchungen über das Wachstum der Maispflanze. Landw. Jahrb. 1882. p. 278.

nesium kommen, wie in Schösslingen, hauptsächlich in jungen procambialen Elementen vor, die vielleicht ihre Wanderbahn herstellen. Im Urmeristem fehlen nachweisbare Mengen von Schwefel und Chlor. Die mehr als 40 cm lang gewachsenen Wurzeln zeigen ziemlich starke Nitratreaction an der Rinde, welche von von aussen aufgenommenen Salzen herrührt.

In den jüngsten Scheideblattanlagen in der Umgebung des Vegetationspunktes lässt sich sehr früh eine Ansammlung von Phosphor und Magnesium beobachten. Uebrigens bedarf dies keiner Besprechung mehr.

# VII. Ueber die Entleerung der Reservestoffe.

Die Versuche von Hansteen¹) und Puriewitsch²) haben die Thatsache festgestellt, dass bei Endospermen, Samenlappen, Knollen und Rhizomen die Entleerung der deponierten Reservestoffe mehr oder minder selbstthätig stattfinden kann. Nun schien es mir geboten zu bestimmen, erstens inwieweit die Entleerung des Rhizoms unabhängig von wachsenden Schösslingen vor sich gehen kann, und zweitens in welchem Grade die Entwicklung der Schösslinge durch die totale oder partielle Separierung vom Rhizomsystem beeinflusst wird. Zu diesem Zweck habe ich Mitte April eine Anzahl kräftig wachsender, unterirdischer Schösslinge aufgesucht und verschieden tiefe Einschnitte in ihre Stieltheile und benachbarte Rhizominternodien gemacht. Die betreffenden Rhizomtheile waren in diesem Stadium mit Stärke strotzend erfüllt, wie ich durch die Musterung zahlreicher Exemplare überzeugt war; es zeigte sich folgendes:

<sup>1)</sup> Hansteen, Über die Ursache der Entleerung der Reservestoffe aus Samen. Flora. 1894 Ed. 79, p. 419.

<sup>2)</sup> Puriewitsch, Physiologische Untersuchungen über die Entleerung der Reservestoffbehälter. Jahrb. f. wiss. Bot. Bd. XXXI, p. 1.

	Stärke	Glykose	Rohrzucker
Rindenparenchym	strotzend erfüllt (5)	sehr wenig	ziemlich viel
Centralcylinderparen- chym	strotzend erfüllt- recht viel (5-4)	desgl.	desgl.
Markparenchym	strotzend erfüllt (5)	desgl.	desgl.

In der folgenden Tabelle stelle ich die Ergebnisse einiger Versuche zusammen:

			Opera-	Inhalt der Rhizome		Bemer-
			tionen	Stärke	Zucker	kungen
I.	operiert : nntersucht :	20. April 30. Mai	Durchschneiden im 6sten Inter- nodium vor und im 3ten Int. hinter dem Schösslinge; Durchschneiden im Stiel	Rinden- und Centralcylinder- parenchym: keine Stärke (0); Mark- parenchym: wenig Stärke (2)	Rinden-, Cen- tralcylinder- und Mark- parenchym: fast kein Zucker	Zuwachs des Schösslings =0
II.	operiert : untersucht ;	20. April 30. Mai	wie oben	wie oben	Rinden-, Cen- tralcylinder- und Mark- parenchym: wenig Zucker	Zuwachs des Schösslings =0
III.	operiert : untersucht :	16. April 18. Mai	Durchschneiden im 3ten Int. vor dem Schösslinge; Durchschneiden im Stiel	keine Stärke (0)	keine Glykose; kein Rohr- zucker	Zuwachs des Schösslings =0
IV.	operiert : untersucht :	16. April 18. Mai	Durchschneiden im 3ten Int. vor dem Schösslinge; 1.5em tiefer Ein- schnitt ins 2ten Int. hinter dem Schösslinge	keine Stärke (0)	keine Glykose; sehr wenig Rohrzucker	Zuwachs des Schösslings =2.5 cm
V.	operiert : untersucht :	20. April 18. Mai	Durchselmeiden im Stiel	fast keine Stärke (0-1)	keine Glykese	Zuwachs des Schösslings =0
VI.	operiert : untersucht :	<ul><li>16. April</li><li>19. Mai</li></ul>	Durchschneiden im 3ten Int. vor dem Schö-slinge	wenig Stärke (2)	fast keine Glykose	Zuwachs des Schösslings =10 cm
VII.	operiert : untersucht :	16. April 19. Mai	Durchschneiden im 2ten Int. hinter dem Schösslinge	keine Stärke (0)	wie oben	Zuwachs des Schösslings =0

Alle oben angeführten Versuche ergaben übereinstimmend, dass die Entleerung, zumal die Stärkeauflösung in bestimmten Rhizompartien unabhängig von Schösslingen vor sich gehen kann. Puriewitsch hat bei den Versuchen mit den Rhizomen von Curcuma und Rudbeckia gezeigt, dass in solchen Rhizomen eine partielle Entleerung selbstthätig stattfand, wenn die continuierliche Ableitung der Lösungsproducte mittelst Gyps besorgt wurde<sup>1</sup>). Nun in meinen Versuchen war die Bedingung für derartige Stärkeentleerung besonders günstig. Die zahlreichen kräftigen Wurzeln an Rhizomknoten erzeugten zur Zeit einen ansehnlichen Blutungsdruck und der zuckerhaltige Blutungssaft wurde immer fort von den Schnittflächen der Rhizome ausgeschieden. Damit wurde eine fortwährende Wegführung der Lösungsproducte erzielt, welche eine so volkommene Entleerung herbeiführte.

Ferner ist es aus obigen Versuchen ersichtlich, dass die Entwickelung der Schösslinge durch jeden operativen Eingriff in benachbarte Rhizominternodien—d.h. durch jede Herabsetzung des Blutungsdrucks—bald sistiert wird.

Macht man in der Nacht oder frühmorgens ein Bohrloch in ein beliebiges Internodium des kräftig wachsenden Schösslings, so quillt bald ein zuckerhaltiger klarer Saft hervor. Am 19. Mai wurde der Blutungssaft von einem mittleren Internodium eines ca. 1.5 Meter hohen Schösslings von *Phyllostachys puberula* gesammelt. In 100 ccm dieser Flüssigkeit fand ich 0.289 gr Glykose. Ausserdem enthält der Blutungssaft eine kleine Menge der Amide, da er nach Beseitigung des Eiweisses<sup>2</sup>) eine starke Trübung beim Zusatz von Quecksilberoxydnitrat giebt.

<sup>1)</sup> Puriewitsch, l.c. p. 28.

<sup>2)</sup> Nach der Stützer'schen Methode.

Schröter') schrieb: "Später, in den ausgewachsenen Gliedern, findet sich oft ein klares Wasser, das in manchen trockenen Gegenden den Reisenden ein höchst willkommener Fund ist." Derartige mit Wasser erfüllte Markhöhlen habe ich manchmal bei jungen Halmen von Phyllostachys mitis gefunden. Das Wasser ist nichts Anderes als der Blutungssaft, der sich von radialen Rissen an der Peripherie des Diaphragms ausgeschieden hat. einem Falle betrug der Glykosegehalt der Flüssigkeit, die sich in der unteren Internodialhöhle von Phyllostachys mitis befand, 0.269%. Wenn man frühmorgens einen Bambusbusch besucht, so wird man einen förmlichen Regen von Wassertropfen aus den Scheideblattspitzen der wachsenden Schösslinge bekommen.2) Dies in bekannter Weise von Blattspitzen ausgeschiedene Wasser enthält auch Glykose neben einer Spur von Amiden. Eine Zuckerbestimmung der am 28. April gesammelten Flüssigkeit ergab 0.0958% Glykose.

Alle diese Thatsachen weisen darauf hin, dass eine erhebliche Menge der Kohlehydrate und vielleicht auch Amide mit dem Blutungssaft den Schösslingen zugeführt werden. Dadurch werden die Schösslinge mit den Baustoffen genügend rasch versorgt.<sup>3</sup>) Die oben erörterten Bauverhältnisse der Stieltheile lassen sich auch nicht anders denken, als dass hier der ausgiebige Stofftransport nur durch die Bündel und zwar durch die wohl ausgebildeten Gefässe geschehen kann.

<sup>1)</sup> Schröter, Der Bambus und seine Bedeutung als Nutzpflanze. p. 14; Cohn, Über Tabaschir, p. 375.

<sup>2)</sup> Mollisch, Über das Bluten tropischer Holzgewächse. Ann. d. Jard. Bot. Buit. 1898. Suppl. II, p. 23.

<sup>3)</sup> Vergleiche hierzu: Strasburger, Ban und Verrichtungen der Leitungsbahnen. p. 877; Fischer, Beiträge zur Physiologie der Holzgewächse. Jahrb. f. wiss Bot. Bd. XXII, p. 75; p. 150.

# VII. Zusammenfassung.

In Obigem habe ich die wesentlichen Züge der Wachstumsgeschichte der Bambusgewächse darzustellen versucht. Die Hauptresultate werden hier kurz in folgenden Worten zusammengefasst:

- 1. Die Stärke wird in parenchymatischen Zellen der Rhizome, Halme und Wurzeln als Hauptreservestoffabgelagert. Die Verminderung derselben im Winter wurde nicht beobachtet, während zur Zeit des raschen Austreibens von Schösslingen eine unverkennbare Stärkezunahme (transitorisch) in benachbarten Rhizomtheilen constatiert wurde.
- 2. Die Glykose dient als Baumaterial in wachsenden Theilen der Schösslinge und ist in schon fertig gestreckten Internodien derselben transitorisch reichlich aufgespeichert.
- 3. Der Rohrzucker tritt als das Lösungsproduct der Stärke im Parenchym der Rhizome und Halme auf.
- 4. In schnell wachsenden Schösslingen fand eine ausgiebige Eiweisszersetzung statt, dabei trat Tyrosin in bedeutender Menge auf.

Tyrosin und Asparagin zeigen einen weitgehenden Unterschied in ihrem Verhalten. Tyrosin wird schwerer und langsamer für Eiweissregeneration verbraucht, so dass es in schon erwachsenen Theilen eine Zeit lang zurückbleibt. Hingegen ist Asparagin leicht und rasch dazu verwendet und kommt nur an Stellen vor, wo eine lebhafte Stoffbildung stattfindet.

- 5. Gerbstoffe kommen nur in Schösslingen einzelner Arten vor, und Fette spielen hierbei keine wichtige Rolle sowohl als Wanderstoffe wie als Reservestoffe.
  - 6. Phosphor, Kalium, Magnesium und Chlor werden

in den Reservestoffbehältern aufgespeichert, dabei kommt Magnesium vorwiegend in Siebröhren vor. Calcium und Schwefel sind gewöhnlich nicht direct nachweisbar.

- 7. Die Mineralstoffe wandern bei rascher Entwicklung der Schösslinge schnell von den Rhizomen aus und werden in den wachsenden Theilen angesammelt. In der Spitze der Halme, Rhizome und Wurzeln befinden sich Phosphor und Magnesium in direct nachweisbarer Form fast ausschliesslich in Procambialsträngen. Schwefel wird erst im wachsenden Theile der Schösslinge deutlich nachweisbar.
- 8. Die vom Boden aufgenommenen Nitrate werden wahrscheinlich schon in den Wurzeln und Rhizomen zu organischen Verbindungen verarbeitet.
- 9. Die Auflösung der Stärke und die Entleerung der Lösungsproducte aus den Rhizomen können unabhängig von der Entwickelung der Schösslinge fortgehen.
- 10. Der ausgiebige und schnelle Stofftransport nach wachsenden Schösslingen von den Rhizomen kann in Wasserbahnen geschehen. Dafür sprechen vor allem die Blutungserscheinungen der Rhizome und Schösslinge und die Bauverhältnisse der Schösslingsstiele.

Juni 1890.

Botanisches Institut Kaiserl. Universität zu Tokio.



# Verzeichniss der untersuchten Arten.<sup>1</sup>)

Phyllostachys mitis Rivière. (Nom. Jap. Moso-chiku.)

Phyllostachys bembusoides Sieb. et Zucc. (Nom. Jap. Ma-dake.)

Phyllostachys bambusoides Sieb. et Zucc. var. aurea Makino. (Nom. Jap. Hotei-chiku.)

Phyllostachys puberula Munro. (Nom. Jap. Ha-chiku.)

Phyllostachys puberula Munro, var. nigra. (Nom. Jap. Kuro-chiku.)

Phyllostachys Kumasasa Munro. (Nom. Jap. Okame-sasa.)

Arundinaria japonica Sieb. et Zucc. (Nom. Jap. Ya-dakc.)

Arundinaria Simoni Rivière. (Nom. Jap. Me-dake.)

Arundinaria Matsumuræ Hackel. (Nom. Jap. Kan-chiku.)

Arundinaria quadrangularis Makino. (Nom. Jap. Shikaku-dake.)

Arundinaria Hindsii Munro. (Nom. Jap. Kansan-chiku.)

Arundinaria Hindsii Munro, var. graminea Bean. (Nom. Jap. Taiminchiku.)

Arundinaria Fortunei Rivière. (Nom. Jap. Chigo-sasa.)

Arundinaria variabilis Makino. (Nom. Jap. Ne-sasa.)

Arundinaria pygmæa Mitf. (Nom. Jap. Oroshima-chiku.)

Arundinaria Narihira Makino. (Nom. Jap. Narihira-dake.)

Arundinaria Tootsik Makino. (Nom. Jap. To-chiku.)

Bambusa borealis\* Hackel. (Nom. Jap. Suzu-dake.)

Bambusa palmata\* Marliac. (Nom. Jap. Chimaki-sasa.)

Bambusa Veitchii\* Carrière. (Nom. Jap. Kuma-sasa.)

Bambusa paniculata\* Makino. (Nom. Jap. Nemagari-dake.)

Bambusa nipponica\* Makino. (Nom. Jap. Miyako-sasa.)

Bambusa ramosa\* Makino. (Nom. Jap. Azuma-sasa.)

<sup>1)</sup> Die ausführliche Beschreibung der hier angeführten Arten findet man bei Makino, Bambusacee Japoniere (The Botanical Magazine, Vol. XIV, Nr. 156, p. 20 ff.), die beigefügten japanischen Namen sollen zum Herausfinden der betreffenden Arten in der genannten Schrift dienen.

Die mit \* bezeichneten Arten gehören meiner Ansicht nach nicht eigentlich zu Bambusa, sondern sie würden vielleicht eine selbständige Gattung bilden. (Vergl. oben p. 446 und auch Makino, l.e. p. 20).

An dieser Stelle spreche ich Herrn Makino für die von ihm gütigst vorgenommene Bestimmung einiger Arten und auch Herren Asō und Inami für ihre freundliche Unterstützung bei einigen analytischen Arbeiten meinen besten Dank aus.

## K. SHIRATA:

Bumbusa vulgaris Wendl. (Nom. Jap. Daisan-chiku.)
Bambusa nana Roxb. (Nom. Jap. Howo-chiku.)
Bambusa nana var. normalis Makino. (Nom. Jap. Taiho-chiku.)
Bambusa stenostachya Hackel. (Nom. Jap. Shi-chiku.)
Dendrocalamvs latiflorus, Munro. (Nom. Jap. Ma-chiku.)

# Inhalt.

I.	Einleitung	427
II.	Untersuchungsmaterial und Methodisches	429
III.	Die Bauverhältnisse	433
IV.	Der Entwicklungsvorgang der Schösslinge	453
v.	Verhalten der Baustoffe während der Entwicklung	
	der Schösslinge	458
VI.	Ueber Entleerung der Reservestoffe	489
VII.	Zusammenfassung	493

# Erklärung der Tafeln.

### Tafel XXII.

- Fig. 1. Zwei neben einander stehende Siebrörenglieder mit zahlreichen Siebtüpfeln (s./p) an den Seitenwänden, aus Rhizomknoten von Phyllostachys mitis. spl Siebplatte, stp Siebtüpfel. Vergr. 360.
- Fig. 2. Querschnitt durch das Rhizom von Bambusa nipponica. R Rinde, Brg subcorticaler Bastring, cent Centralcylinderparenchym. Vergr. 30.
- Fig. 3. Querschnitt durch das Rhizom von Arundinaria japonica. B Bastbänder. Vergr. 30.
- Fig. 4. Die spindelförmige Auschwellung des Leptoms eines Knospenbündels bei der Ansatzstelle an der Rhizombündel. S Siebröhren, gl Geleitzellen, G Gefässe, P Parenchymzellen, cbf cambiformartige Elemente Vergr. 70.
- Fig. 5. Querschnitt durch die Anschwellung. B Bastzellen, P u. cbf wie in Fig. 4. Vergr. 125.
- Fig. 6. Theil der langgestreckten cambiformartigen Elemente aus dem mittleren Theile der Anschwellung, mit Querstreifen auf den Seitenwänden. Vergr. 450.
- Fig. 7. Derselbe im Querschnitt. Vergr. 450.
- Fig. 8. Die Leptomanschwellung in einem früheren Entwicklungsstadium. Längsschnitt durch den Knoten. S Siebröhren, B Bastzellen, chf cambiformartige Elemente. Vergr. 83.
- Fig. 9. Übergangsstelle der cambiformartigen Elemente zum normal gebauten Leptom, in einem jugendlichen Zustand. Sämmtliche Elemente mit auffallend grossen Zellkernen und reichlichem Plasmagehalt. S. u. cbf wie in Fig. 8. Vergr. 450.
- Fig. 10. Dergleichen im fertigen Zustand. tp Tüpfel, chf wie oben. Vergr. 450. Figuren 4-10 beziehen sich auf Phyllostachys mitis.
- Fig. 11. Ein Gefässbündel aus einem inneren Teil des Rhizoms von Arundinaria Hindsii. S Siebröhren, gl Geleitzellen, G Gefässe, d Durchlassstelle. Vergr. 125.
- Fig. 12. Eine subepidermale sclerotische Parenchymschicht des Rhizoms von Bambusa palmata. Längsschnitt. ep Epidermis, scl sclerotische Parenchymzellen, R Rindenzellen. Vergr. 360.

- Fig. 13. Querschnitt durch den Stieltheil. R Rinde, B Bastbänder, bs Bastscheide des Mestombündels, mes Mestom. Vergr. 17.
- Fig. 14. Ein Mestombündel im Stieltheile, mit vollkommen umschliessender Bastscheide. t Tracheiden, P, bs, S, gl, u. G wie oben. Vergr. 125. Fig. 13-14. Phyllostachys mitis.
- Fig. 15. Querschnitt durch den dünnen Halmzweig von Arundinaria pygmæa. ep u. R wie oben. Vergr. 83.
- Fig. 16. Theil desgleichen von Arundinaria japonica. ep, B u. R wie oben. Vergr. 360.
- Fig. 17. Ein Halm-Bündel von Bambusa nana var. normalis, mit Parenchymlamelle im innenseitigen Bastbeleg. par. l Parenchymlamelle, S. G u. B wie oben. Vergr. 200.
- Fig. 18. Einige verschiedenartige Vorkommnisse des Parenchymgewebes im Bastbelege. Arundinaria Hindsii. B Bastbelege, par parenchymatisches Gewebe. Vergr. 70.
- Fig. 19. Ein Halmbündel von Bambusa nana. Die durch parenchymatische Zellen vom Mestom abgetrennte Masse des Bastbelegs bleibt unverdickt. par. l, B wie oben. Vergr. 200.
- Fig. 20. Auftreten der Stärkekörner in neu differenzierter Parenchymlamelle. Vergr. 70.
- Fig. 21. Obiges im Längsschnitt. Vergr. 125.
- Fig. 22. Die durch successive Quertheilungen von Procambialzellen entstandenen Parenchymzellen. pro Procambialzellen, k Kern, st Stärkekörner. Vergr. 360. Figuren 20-22. Arundinaria Hindsii.



## Tafel XXIII.

- Fig. 23. Querschnitt durch die junge Wurzel von Bambusa palmata. i.R innere Rindenzellen, end Endodermis mit Caspary'schen Streifen, per Pericambium, p.had peripherische Hadromstränge (primordiale Netztracheiden), p.lep peripherische Leptomstränge. Vergr. 360.
- Fig. 24a. Peripherischer Theil der Wurzelrinde von Phyllostachys mitis. ep Rest der Epidermis, hyp stark verdickte (an den Aussenwänden) subepidermale Zellen, sel peripherische sclerotische Elemente, a.R äussere Rindenzellen. Vergr. 200.
- Fig. 24b. Desgleichen im jugendlichen Zustand. cp, hyp u. scl wie oben.
- Fig. 25. Starkverdickte Subepidermalzellen (Aussenscheide) von *Phyllostachys bambusoides* var. aurea. Vergr. 360.
- Fig. 26. Peripherischer Theil der Wurzelrinde von Bambusa vulgaris. ep Epidermis, hyp unverdickt gebliebene Subepidermalzellen, scl peripherische Sclerenchymzellen, a.R äussere Rindenzellen. Vergr. 360.
- Fig. 27. Wurzelrinde von Bambusa nana. ep, hyp, scl, a.R u. i.R wie oben. Vergr. 125.
- Fig. 28. Endodermis (end) und starkverdickte Pericambiumzellen (per) von Phyllostachys mitis. Verg. 360.
- Fig. 29. Längsschnitt durch die Endodermis von *Phyllostachys mitis*. Vergr. 360.
- Fig. 30. Längsschnitt durch den peripherischen Theil der Wurzehrinde von Arundinaria Matsumuræ. hyp u. scl wie in Fig. 24. Vergr. 360.
- Fig. 31. Endodermis und angrenzende Rindenzellen von Bambusa stenostachya. Längsschnitt, cel Zellstoffauswüchse, end u. per wie oben. Vergr. 360.
- Fig. 32. Dieselben im Querschnitt. end, per u. cel wie oben. Vergr. 360.
- Fig. 33. C-förmig verdickte Endodermiszellen von Bambusa palmata. Vergr. 360.
- Fig. 34. Theil des Wurzelquerschnittes von *Phyllostachys Kumasasa*. ver Verstärkungsring, *If* Lufträume, *end*, *i.R*, *per* u. *p.lep* wie oben. Vergr. 360.
- Fig. 35. Verknüpfung der Leptomstränge durch dünnwandiges Verbindungsgewebe, Phyllostachys bambusoides. ver.p. Verbindungsgewebe, nb.w Nebenwurzel, p.lep, i.lep, G, end wie oben. (schematisiert) Vergr. 70.

- Fig. 36. Dergleichen bei Bambusa vulgaris. Vergr. 45.
- Fig. 37. Querschnitt durch die Hauptwurzel an der Ansatzstelle der Nebenwurzel. Arundinaria Matsumuræ. n.lep Leptomstrang der Nebenwurzel, lep Leptomstränge der Hauptwurzel, end, i.R, per wie oben.
- Vergr. 360. Fig. 38. Innerer Leptomstrang von *Bambusa vulgaris*. S Siebröhre, cb Cambiformzellen, mz mechanische Zelien. Vergr. 360.
- Fig. 39. Verschmelzung des inneren Leptomstrangs mit dem peripherischen.

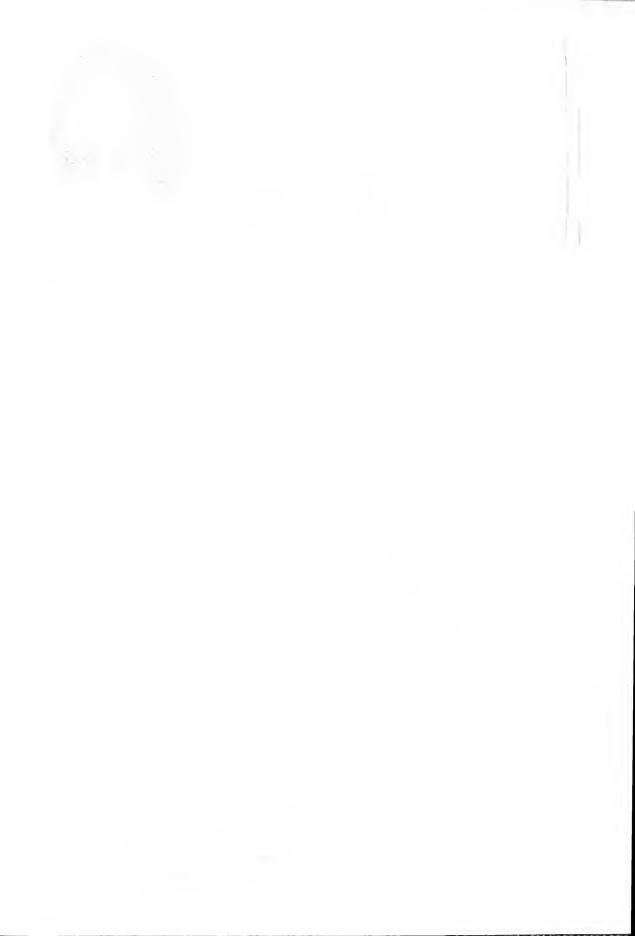
  i.lep innerer Leptomstrang, p.lep peripherischer Leptomstrang, S,
  cb wie oben. Vergr. 360.
- Fig. 40. Verschmelzung zweier peripherischen Leptomstränge. Vergr. 360.
  Fig. 41. Directer Anschluss des Leptomstrangs an Hadromparenchym. Gefäss, hp Hadromparenchym (Gefässbelegzellen), lep u. mz wie oben.
- Vergr. 360. Fig. 42. Zusammentreffen zweier Hadromstränge. G, hp u. mz wie oben. Vergr. 360.
- Figuren 39-42. Phyllostachys bambusoides.

  Fig. 43. Verbindungsgewebe zwischen inneren und peripherischen Leptomsträngen. i.lep, p.lep, mz u S wie oben. Vergr. 360.
- Fig. 44. Dasselbe von Bambusa vulgaris im Längsschnitt. S, mz, ver.p wie oben. Vergr. 360.
  Fig. 45. Querschnitt durch den Basaltheil der Nebenwurzel von Phyllostachys
- mitis. lep Leptomstränge, mz mechanische Zellen. Vergr. 360.
  Fig. 46. Theil der Rinde der Nebenwurzel von Phyllostachys puberula, mit endophytischen Mycelfäden. R Rindenzellen, myc Pilzfäden, ves Vesiculen, kor gelbe körnige Substanz. Vergr. 360.
- Fig. 47. Querschnitt durch die Nebenwurzel von *Phyllostachys puberula*.

  hyp Subepidermalzellen, scl peripherische sclerotische Zellen, R, myc, end, lep wie oben. Vergr. 200.

#### Tafel XXIV.

- Fig. 48. Ein Scheideblattbündel mit starkem Bastbeleg (B) von *Phyllo-stachys mitis*. Vergr. 83.
- Fig. 49. Querschnitt durch das Scheideblatt von Arundinaria Matsumuræ Vergr. 360.
- Fig. 50. Kleinere Scheideblattbündel von *Bambusa stenostachya*; Leptom (*lep*) ist stets von einschichtigen verholzten Elementen (*h*) umgeben. Vergr. 360.
- Fig. 51. Querschnitt durch das Scheideblatt von Bambusa stenostachya. B Bastbelege, subcp.b subepidermale Bastplatte, P Parenchym, mes Mestom. Vergr. 83.
- Fig. 52. Desgleichen von Phyllostachys mitis. lf Lufträume, B, subep. b, P, mes wie oben. Vergr. ca. 10.
- Fig. 53. Queranastomose des Scheideblattbündels von Arundinaria Hindsii. Vergr. 360.
- Fig. 54. Ligninauswüchse an Zellwänden. ("Zwickel"). Vergr. 360.
- Fig. 55. Ein kleines Bündel im Laubblatt von Arundinaria Hindsii. ps Parenchymscheide, bs Bastscheide, subep.b, Had, Lep wie oben. Vergr. 360.
- Fig. 56. Längsschnitt durch einen kleinen Nerv des Laubblattes von Bambusa palmata. ep, subep. b, ps, bs wie oben. Bastscheideelemente sind reichlicher betüpfelt als subepidermale Bastelemente. Vergr. 360.
- Fig. 57a. Stärkekörner aus Rhizom von Phyllostachys Kumasasa. Vergr 830.
- Fig. 57b. Polyadelphische Stärkekörner aus dem Halm von Bambusa palmata. Vergr. 830.
- Fig. 58. Einige Tyrosinkrystalle, die ausserhalb des nach Borodin'scher Methode behandelten Schnittes entstanden sind. Vergr. 360.
- Fig. 59. Beim Schneiden sofort in jungen Bastzellen auskrystallisierende Tyrosinkrystalle. B junge Bastzellen (mit plasmatischem Wandbelege), P Parenchymzellen, tyr Tyrosinkrystalle. Vergr. 360.
- Fig. 60. Desgleichen in Längsschnittansicht. Vergr. 200.
- Fig. 61. Beim Einlegen vom Schnitt in Glycerin in das Zelllumen ausgeschiedene Tyrosinkrystalle. *tyr* Tyrosinkrystalle, *st* Stärkekörner, *P*. Parenchymzellen. Vergr. 360.





# Decomposition of Hydroxyamidosulphates by Copper Sulphate.

Bv

Edward Divers, M. D., D. Sc., F. R. S., Emeritus Prof.,

and

Tamemasa Haga, D. Sc., F. C. S., Professor, Tökyö Imperial University.

When copper sulphate is added to a solution of a hydroxy-amidosulphate and the mixture heated, the acid of the salt is quickly decomposed into water, sulphur dioxide, sulphuric acid, amidosulphuric acid and nitrous oxide, with possibly a little nitrogen. By itself, a heated solution of an alkali hydroxy-amidosulphate is in a state of very unstable equilibrium, generally hydrolysing into a solution of hydroxylamine acid sulphate, and always doing so in presence of a trace of acid, whilst in presence of even a trace of alkali it slowly passes into sulphite and hyponitrite (this Journ, 3, 219). In the cold with alkali and copper salt, the hydroxyamidosulphate becomes oxidised at once to sulphite, sulphate, nitrous oxide, and water with reduction of the cupric hydroxide (op. cit., 225), and when heated with cupric chloride it reduces the latter to cuprous

chloride, becoming itself converted into sulphur dioxide, sulphate, nitrous oxide, and water. Mercuric nitrate oxidises hydroxyamidosulphate more completely, but ferric chloride seems to act like copper sulphate, and liberates sulphur dioxide.

An alkali hydroximidosulphate is also decomposed by copper sulphate, but not so easily, for it can be heated with it at 100° for a short time without change, and only decomposes (but then suddenly) some degrees above that temperature, yielding the products which a hydroxyamidosulphate gives, together with sulphuric acid from its hydrolysis into that salt.

Although the presence of much sulphuric acid prevents the action of copper sulphate on a hydroxyamidosulphate, the acid in moderate excess has but little effect.

Sodium hydroximidosulphate, if kept with care, decomposes only very slowly in a way which has hitherto been obscure (this Journ, 7, 45), but if considered in connection with the action of copper sulphate it may be regarded as essentially the same as that brought about by heating it in solution with that salt. For, the decomposed hydroximidosulphate contains, besides acid sulphate and hydroxyamidosulphate, both a little gas (nitrous oxide or nitrogen) shut up in its pores which escapes when the mass is dissolved in water, and also a little amidosulphate, which can be separated from the other salts by precipitation with mercuric nitrate (this Journ., 9, 242, also 229, 230).

The decomposition of hydroxyamidosulphates by copper sulphate is also in evident relation with the gradual decomposition of impure hydroxylamine hydrochloride, particularly when ferric chloride is among the impurities, water, nitrous oxide and ammonia (in place of amidosulphuric acid) being the principal, if not the sole, products.

There is a very marked difference in the proportions of the products of decomposition between a hydroxyamidosulphate and a hydroximidosulphate, but this seems to be owing merely to the fact that the temperature of the decomposition is different, for according as hydroxyamidosulphate is heated slowly or rapidly the proportions of the products of decomposition deviate from or approach those which obtain when a hydroximidosulphate is decomposed, this only taking place at a temperature above 100°.

As little as one-tenth of an equivalent of copper sulphate has been found to suffice for the complete decomposition of an alkali hydroxyamidosulphate, the copper sulphate not being consumed in the change it effects; this allows of the decomposition being to a great extent carried out at the boiling temperature, when again the result approaches that observed where hydroximidosulphate is the salt decomposed. Even much less than the amount above named will effect an almost complete decomposition but that the quantity of the catalytic agent cannot be very greatly reduced seems to be due in part to the simple hydrolysis of some of the hydroxyamidosulphuric acid set free by the copper sulphate during the prolonged hereing here necessary.

Since the cupric salt suffers no reduction, it will be seen that one part of the hydroxyamidosulphate becomes reduced to amidosulphate by yielding oxygen for the oxidation of the other part to water, sulphate and nitrous oxide. The following equation shows that the hydroxyamidosulphate may change by cumulative resolution, half into a reduced product (amidosulphate), and half into oxidised products together equivalent to the non-existent dihydroxyamidosulphate:

(1). 
$$2Cu(H_2NSO_4)_2 = N_2O + H_2O + H_2SO_4 + CuSO_4 + Cu(H_2NSO_3)_2 = Cu(H_2NSO_5)_2 + Cu(H_2NSO_3)_2.$$

Such an equation expresses much of what happens in the decomposition of a hydroxyamidosulphate at a lower temperature, but even in this case, and much more in the decomposition of a hydroximidosulphate by copper sulphate, where the temperature is higher, a third molecule decomposes in another way. The result is that the free sulphuric acid shown in the above equation gets neutralised, and the third molecule of hydroxyamidosulphuric acid yields neither sulphate nor amidosulphate, all its sulphur being eliminated as dioxide, its nitrogen as nitrous oxide, and its hydrogen as water thus reverting to sulphurous and hyponitrous acids, just as it does under the influence of an alkali (p 497) adding to equation (1) that of  $Cu(H_2NSO_4)_2=N_2O+2H_2O+2SO_2+CuO$ , we get (2),  $3Cu(H_2NSO_4)_2=2N_2O+4H_2O+2SO_2+2CuSO_4+Cu(H_2NSO_3)_2$ , with products free from acid.

It is possible to express the decomposition of hydroxy-amidosulphate differently, by making nitrogen one of the products in place of nitrous oxide, thus:

$$(3) \ 3Cu(H_2NSO_4)_2 = 2N_2 + 2H_2O + 2H_2SO_4 + 2CuSO_4 + Cu(H_2NSO_3)_2;$$

(4) 
$$Cu(H_2NSO_4)_2 = N_2 + 2H_2O + SO_2 + CuSO_4.$$

In (3) sulphur dioxide is not a product whilst in (4) it is. Whether, however, nitrogen is formed, even in small quantity, is doubtful. Along with the nitrous oxide soluble in alcohol, we found a little insoluble gas—about 4 per cent. by volume of the whole gas,—but we are not prepared to assert that this was not due to air in spite of the precautions we took to expel all air from the apparatus by carbon dioxide before the decomposition. It will be seen from the equations that, with nitrous oxide as a product of the decomposition, the sulphur appearing as sulphate

equals that as amidosulphate, whereas, with nitrogen as a product, the sulphur as sulphate is double that as amidosulphate in (3), whilst in (4) there is none as amidosulphate. Now, in the observed decompositions of hydroxyamidosulphate the sulphur as sulphate has been found equal, on the average, to that as amidosulphate, a result showing that within the limits of accuracy of the somewhat complex analytical work, no nitrogen is generated.

Although, when using copper sulphate or copper hydroxy-amidosulphate, no change to cuprous salt is observable, the reduction of cupric chloride to cuprous chloride points clearly to the activity of the copper salt as a 'carrier of oxygen' from one molecule of the hydroxyamidosulphate to another.

## Results and Method of the Quantitative Experiments.

The results of the experiments are given, not in the order in which they were obtained but in that of the growth in quantity of the sulphur dioxide produced.

In an experiment in which copper hydroxyamidosulphate was heated very slowly, so as to carry out the decomposition at as low a temperature as possible (boiling the solution only at the end in order to expel the last portions of sulphur dioxide), results were obtained which agree sufficiently well with those calculated on the assumption that 3.7 per cent. of the salt gives all its sulphur as dioxide, its hydrogen as water, and its nitrogen as nitrous oxide, whilst the rest of the salt decomposes according to equation (1):

Sulphur as dioxide; as trioxide and amidosulphate.

Found3.5	96.2
Calc3.7	96.3

An experiment with sodium hydroxyamidosulphate and its equivalent of copper sulphate, gave results indicating that about 5.3 per cent yielded all its sulphur as dioxide, the rest of the salt giving sulphur trioxide (sulphuric acid) and amidosulphate (equation 1):

Sulphur as dioxide; as trioxide; as amidosulphate; as acidity.

Found5.5	46.0	48.0	21.6
Cale5.3	47.4	47.4	21.0

Copper hydroxyamidosulphate in four experiments gave results agreeing nearly with the assumption that 13.2 per cent. of the salt gave sulphur dioxide, the rest decomposing according to equation (1):

Sulphur as dioxide; as trioxide; as amidosulphate; as acidity.

Found13.0	43.0	43.6	11.1
,,13.0	43.3	43.2	
"   13.1	S	6.6	
,,13.3	8	6.5	
Calc13.2	43.4	43.4	15.1

In another experiment copper hydroxyamidosulphate gave the following results, as against calculation for 15.4 per cent. to decompose so as to yield its sulphur as dioxide:

Sulphur as dioxide; as trioxide; as amidosulphate; as acidity.

Found15.1	42.9	41.6	10.7
Cale15.4	42.3	42.3	13.5

In one more trial, copper hydroxyamidosulphate decomposed nearly as if 16.6 per cent. of it yielded all of its sulphur as dioxide:

Sulphur as dioxide; as trioxide and amidosulphate; as acidity.

Found16.3	83.3	9.3
Calc16.6	83.4	12.5

A solution of potassium hydroximidosulphate heated with very little more than its equivalent of copper sulphate, gave results showing that 25 per cent. of the salt yielded all the sulphur of the hydroxyamidosulphate coming from it by hydrolysis, as sulphur dioxide:

Sulphur as dioxide; as trioxide; as amidosulphate; as acidity.

Found25.2	37.3	37.0	5.5
Cale25.0	37.5	37.5	6.25

A solution containing sodium hydroximidosulphate and copper sulphate decomposed in two experiments, in such a way that about 28 per cent. of the hydroxyamidosulphate sulphur became dioxide:

Sulphur as dioxide; as trioxide; as amidosulphate; as acidity

Found 27.6	36.9	35.0	4
,,28.0	36.2	35.8	5.8
Calc 28.0	36.0	36.0	4

The numbers in the above table stand for parts per hundred of the sulphur of the total hydroxyamidosulphate decomposed, and not of the sulphur of the hydroximidosulphate even when such a salt has been that experimented with. The 'acidity' sulphur is calculated as if the acidity is due to sulphuric acid, not amidosulphuric acid. The 'trioxide' sulphur is that of the sulphuric acid and copper sulphate yielded by the decomposition. The differences between the calculated quantities and those found must be largely attributed to imperfect estimation; they cannot be due to error in theory, because no other explanation of the change than that adopted is possible. In a copper-salt

solution mixed with much barium sulphate, it was not easy to titrate acid with lacmoid paper as indicator. The separation of sulphate and amidosulphate is not a simple process, especially when much sulphate is present derived from sources other than the reaction to be dealt with.

The salt employed in the experiments was either copper hydroxyamidosulphate, or sodium hydroxyamidosulphate with copper sulphate, or one of the alkali hydroximidosulphates with copper sulphate.

- 1. A solution of the copper salt, containing only a very little copper sulphate was prepared from normal barium hydroxyamidosulphate and copper sulphate, the barium salt (this Journ., 3, 213, 216) had to be prepared as wanted, because of the instability of the hydroxyamidosulphates. The strength of the solution was determined by a barium estimation (hydrolysis in sealed tube and weighing of the barium sulphate). Copper sulphate in slight excess and carefully weighed was added to the weighed solution of the barium salt, and the copper hydroxyamidosulphate at once used without filtering off the barium sulphate.
- 2. Sodium hydroxyamidosulphate solution was prepared just before use by hydrolysing a centigram-molecule of the hydrox-imidosulphate by adding to its solution a minute and known quantity of sulphuric acid (this Journ., II, 3) and to it was added after neutralisation with sodium hydroxide half a centigram-molecule of copper sulphate.
- 3. Potassium or sodium hydroximidosulphate in the quantity of a centigram-molecule was dissolved and directly heated with a half molecule in centigrams of copper sulphate.

The solution (either 1, 2, or 3) being in a small flask

connected with a tube receiver holding bromine water kept cold, was heated, sometimes quickly, sometimes slowly, either by a spirit lamp or in a bath of sulphuric acid, the solution being finally boiled for some minutes, so as to drive all sulphur dioxide into the bromine water. Before heating, air was removed from the apparatus by a current of carbon dioxide. In one experiment the apparatus was made entirely of glass. The oxidised sulphur, dioxide was weighed as barium sulphate.

The boiled-out copper solution was titrated with N/10 soda (free from sulphate), using lacmoid paper as indicator. The imperfection of this operation was proved beyond doubt on calculating out the nature of the changes which had occurred, but it was serviceable and the best available under the circumstances.

To the boiling hot solution and precipitate of barium sulphate, barium chloride was added in excess, the total precipitate collected, well washed, and transferred to a pressure tube in which it was heated with hydrochloric acid for three hours at 150°. The barium sulphate was again washed on the filter, then ignited and weighed. The second filtrate and washings contained sulphuric acid, the quantity of which was estimated as barium salt. To make this part of the analytical process intelligible, it must be explained that barium amidosulphate, although itself quite soluble in water, is partially precipitated along with barium sulphate, even in presence of hydrochloric acid (this Journ., 9, 283). At 150°, the precipitated amidosulphate hydrolyses, yielding barium sulphate and ammonium sulphate in molecular proportions.

The copper filtrate from the crude barium precipitate was evaporated to a small volume, heated with hydrochloric acid for some hours at 150°, and mixed with barium chloride. The precipitated barium sulphate represented the principal quantity

of amidosulphate sulphur, the full amount of which was ascertained by adding to it twice the quantity of that in the ammonium sulphate extracted by hydrolysis from the crude barium precipitate. The sulphur from the hydroxyamidosulphate, obtained as sulphate, was found by subtracting from the total the sum of the quantities of sulphur present as (a) copper sulphate taken; (b) barium sulphate from the hydrolysed barium amidosulphate which had been precipitated along with the barium sulphate by barium chloride; (c) sulphuric acid added for hydrolysing the hydroximidosulphate, when that salt had been started with; and (d) in the same case, sulphuric acid resulting from the hydrolysis of the hydroximidosulphate to hydroxyamidosulphate.

Hardly any attempt was made to estimate the amount of nitrous oxide liberated. To do so would only have been useful as a check on the accuracy of the determinations of the amidosulphate, and for that purpose the two substances would have had to be estimated in the products of one experiment. This, it did not seem possible to do. An experiment in which hydroxyamidosulphate was decomposed gave 55.3 per cent. of the nitrogen as nitrous oxide, as against 56.6 calculated from the equation most in accordance with amidosulphate and other sulphur determinations. The method of measuring the nitrous oxide and nitrogen was to expel air from the apparatus by a current of carbon dioxide continued for some time, and then heat the copper salt and boil out the gases which were collected over mercury and potassium hydroxide and measured. The alkali was then replaced by absolute alcohol to dissolve the nitrous oxide, and the residual gas measured.

# Observations on the Development, Structure and Metamorphosis of Actinotrocha.

By

Iwaji Ikeda, Rigakushi.

With Plates XXV-XXX.

## Introductory.

Since the discovery of Actinotrocha by Johannes Müller in 1846, this peculiar larval form and its mother animal, Phoronis, have been made the subject of investigations by many distinguished authors such as Wagener ('47), Gegenbaur ('54), Krohn ('54), Schneider ('62), Metschnikoff ('72, '82), E. B. Wilson ('81), and Foettinger ('82). Among more recent writers Caldwell ('85), McTntosh ('88), Benham ('88), Roule ('90, '96), Corl ('91), and E. Schultze ('97) may be mentioned as having published important contributions; while Masterman ('97) has made quite an elaborate study of the animal with the view of establishing its relationship to Balano-glossus and the Chordata in general. As, however, in spite of all these works there still existed many gaps and unsatisfactory points

in our knowledge of this interesting animal, the investigation, of which an account is given in the following pages, was undertaken, and though the results are far from exhaustive, I hope they will help to advance our knowledge of the subject.

My study was begun in the summer of 1898 during a stay at the Misaki Marine Biological Station and later was continued at that Station as well as in the Zoological Institute of the Science College.

At Aburatsubo, a small inlet close to the Station, is found a species of *Phoronis*, which has been named by Dr. Oka ('97) *P. ijimai*.\* Its colonies adhere to the overhanging ledges of rocks near the shore. As the water at the place is always calm and at low tides recedes so as to almost expose the ledges, the animals can be easily collected. During the greater part of the year, eggs and young embryos, clustered together, in what may conveniently be called *embryonal masses*, are found adhering to the lophophoral crown of the adult, one on each side of the median line. These furnished materials for the study of fertilization, segmentation and the early larval stages. The larvae in the Actinotrocha stage are found swimming in the inlet and are caught with the surface net. As will later be fully described, there occur four kinds of the larvae, which no doubt represent as many species, including the common *Phoronis ijimai*.

The specimens, both adult and larval, were killed with the saturated solution of corrosive sublimate in 1% acetic acid or with Flenning's fluid. Of the various colouring methods tried on the sections, double-staining with cosin or safranin and Delafield's hamatoxylin gave the most satisfactory results.

<sup>\*</sup>For a discussion of the status of this species, see Snoplementary Notes.

Before proceeding further, I beg to tender my sincere thanks to Professors Mitsukuri and Lima for their kind supervision of my work and for their painstaking revision of my manuscripts.

#### Contents.

- 1. The Early Development of the Phoronis Larva.
  - a. Notes on Fertilization.
  - b. Notes on Segmentation.
  - c. Gastrulation and Mesoblast-Formation.
  - d. Further Observations on the Development of the Luva, up to the Period when it becomes free-swimming.
- II. The Structure of Actinotrocha.
  - a. The External Appearance of Actinotrocha.
  - b. The Internal Structure of Actinotrocha.
    - 1. Body-Divisions and Body-Cavities,
    - 2. Organs of Ectoblastic Origin.
    - 3. Organs of Entoblastic Origin.
    - 4. Organs of Mesoblastic Origin.
- III. Metamorphosis.
- IV. Supplementary Notes.

## I. The Early Development of the Phoronis Larva.

### a. Notes on Fertilization.

Phoronis is, as is well known, a hermaphrodite, in which both the male and female sexual elements mature at nearly the same time. But few authors seem to have studied the animal during its breeding season, so that our knowledge of its sexual organs and, consequently, of its fertilization has remained very imperfect, as was pointed out by Cori ('91). The only existing statement as to how and where fertilization is accomplished in *Phoronis* is that of Kowalewsky ('67). This author thought that fertilization

took place in the body-cavities, and accordingly, as Cori remarks, he must have believed that self-fertilization prevails in *Phoronis*. Cori considers this as highly improbable, but does not bring forward any positive facts in contradiction of it, his inference being drawn solely from facts observed in other marine Metazoa.

Since Kowalewsky's valuable researches ('67), it has generally been accepted that the nephridia serve also as oviducts. Thus Benham says that he saw an ovum attached to one side of the nephridial funnel and further mentions that Kowalewsky observed eggs moving through the nephridial canal towards the exterior. Unfortunately both observers failed to clucidate what stage of development these eggs are in.

In *Phoronis ijimai* mature sexual elements are constantly discovered throughout about one half of the year (from November to May or June). By carefully examining a living colony of that species during this period, it will soon be perceived that some individuals differ slightly from the rest in the aspect of the foot or body. We see in them a moniliform series of small white specks shining through the skin in the appermost part of the body. These are the oval ready to escape to the exterior through the nephridia. It must have been such individuals that were observed by Kowalewsky and Benham. The body-cavity, in which the ova lie, corresponds to the rectal chamber near the anterior end of the body. I have endeavoured to ascertain whether these ova are fertilized or not, and have at last succeeded in ascertaining that they are in a stage prior to the extrusion of polar globules, —the primary occutes, in Bovert's terminology. In the fresh state, they are spherical or somewhat elliptical in shape and perfectly opaque by virtue of the abundant volk-granules contained in the vitellus.—It is characteristic of these ova that the nucleus, which

is situated, not in the centre, but near the periphery, is always in the meta- or ana-phase of Karyokinesis (fig. 17). In such an ovum the chromosomes are constantly found to be six in number, each being dumb-bell shaped with the two ends directed towards the poles. Fig. 18 represents a portion of the section passing through the equatorial plane of the nuclear figure. It is evident that these eggs are in preparation for the extrusion of the first polar globule. As shown in the above figure, the finely granular protoplasm of the vitellus contains thickly and uniformly distributed volk granules, which have a strong affinity for cosin.

That the eggs in question are mature is further demonstrated by the fact that I succeeded in artificially fertilizing them and in rearing out of them normal embryos which grew to certain advanced stages of development.

If we now examine the embryonal masses, which, as has been mentioned, are found attached one on each side of the tentacular crown of the adult *Phoronis*, we find that the embryos which are farthest away from the nephridial pores are the most advanced in development and that they are found in successively younger and younger stages as we approach the pores, until we reach such eggs as have just been fertilized or perhaps even such as have not yet been fertilized at all. But even the youngest eggs found in the mass present an appearance very different from those found in the body-cavities, the former being invariably at a stage after the expulsion of one or two polar globales. In the egg taken from the mass and shown in fig. 19, two polar globules have already been formed; these are situated close together just inside the vitelline membrane.

On the other hand, if we examine by means of serial sections through the posterior region of an adult, where the stomach and

the sexual organs lie grouped together, a number of large eggs are frequently found, floating freely in the coclomic fluid of the body-cavities. These eggs do not differ in any respect from those in the nephridial region as regards the size, the appearance of the karyokinetic figure, or the number of chromosomes.

The facts above stated plainly point to the following conclusion:—The oogonia fall into the body-cavities by a dehiscence of the ovarian walls and here develop until they reach the stage of primary oocytes. These travel gradually upwards to the nephridial region, retaining meanwhile the nuclear figure formed for the production of the first polar globale. Reaching that region, the primary oocytes are destined sooner or later to be carried by way of the nephridia to the exterior, where they become fertilized by spermatozoa from other individuals.

Reserving an account of the spermatogenesis and ovogenesis for a future occasion, I may here refer to a few facts observed by me relative to the process of fertilization. When the two sexual elements are artificially brought together, numberless spermatozoa soon attach themselves to the surface of the ovum. About 10 minutes afterwards, the first polar globule makes its appearance, followed soon afterwards by the second. Meanwhile a small clear spot, probably marking the place where the male element has entered, appears on the surface of the egg; it is however observable for only a very short time. Both figs. 19 and 20 are sections of ova taken from the embryonal mass. The ovum given in fig. 19 is fully mature and ready to be fertilized; close to the polar globules rests the large female pronucleus. The ovum represented in fig. 20 belongs to a stage of fertilization in which the two pronuclei stand closely side by side. The larger female pronucleus has a nuclear membrane irregular in contour. The

intensely stained chromatin pieces are in both nuclei dispersed without any apparent order throughout the finely granular nuclear substance. At one spot outside the male pronucleus, there is visible a small and clear archoplasmic (?) space surrounded by a set of exceedingly fine radial rays. The two polar globules of this egg were distinctly visible in other sections which have not been figured.

### b. Notes on Sugmentation.

Our knowledge of the mode of segmentation in *Phoronis* is far from being satisfactory. Metschnikoff ('82) gives no account of the process. Foethiger ('82), if one may judge from his figures, seems to have seen the egg undergoing to taland unequal segmentation. According to Caldwell ('82), the segmentation "proceeds with considerable regularity" (l.c., p. 374); Roule ('90) says "l'orde fécondé subit une segmentation totale fort regulière....." (l.c., p. 1147). E. Schultze ('97) simply says "Ich sah das Ei sich total und unäqual furchen" (l.c., p. 6).

My observations of the process were made on eggs found in the embryonal mass as well as on those artificially fertilized. As the former showed comparatively rarely the earlier stages of the segmentation, it was necessary to have recourse to the latter for filling up the gaps of observation.

Soon after the formation of the second polar globule and the disappearance of the micropyle-like spot the first cleavage line makes its appearance, passing on one side of the polar globules (figs. 1 and 21). At this stage I can not perceive any difference in size and structure between the two blastomeres. The second

cleavage plane passes at right angles to the first (fig. 2 b). It is a remarkable peculiarity of *Phoronis* eggs that two sister blastomeres derived by the division of a mother blastomere, never undergo the next division simultaneously, so that between any two consecutive stages having an even number of blastomeres there intervenes an intermediate stage with an odd number of the same. This phenomenon occurs even at the second cleavage; thus just before the egg attains the four-cell stage, there exists a stage of three cells, such as is seen in fig. 2 a. Among the later stages, those of 5, 7, 9........cells are of constant occurrence. Consequently it is scarcely admissible to say that the segmentation proceeds with considerable regularity.

Caldwell ('82) has asserted that the first differentation of the future blastoderm into the ectoblast and entoblast is observable as early as in the four-cell stage. He says: "At the stage of 4 segmentation-spheres a division into two smaller clear and two larger opaque cells indicates the future ectoderm and endoderm" (l.c., p. 374). At the corresponding stage of Phoronis *ijimai* I have not been able to discover any appreciable difference in the size of its cells (see fig. 2 b). Following the 4-cell stage, the division of the blastomeres in the equatorial plane puts the egg on the way to the 8-cell stage. According to my own observations, the above mentioned difference in size of the blastomeres becomes first perceptible at this stage. Fig. 3 shows a side view of an egg with 8 blastomeres; it will be seen that the upper four blastomeres are very slightly smaller than the lower four. I could not however, at that period, recognize any difference in the cell-contents of the two classes.

The irregularity of division, which, as before mentioned, becomes more and more pronounced as segmentation advances, tends

to gradually obscure the orderly arrangement of the cells. At the 16-cell stage the regular arrangement is still, though less distinctly, maintained, while at the 32-cell stage it is quite disturbed (fig. 4). From this period on, the polar globules can no longer be detected.

In the earlier stages of segmentation, the blastomeres are found in close contact with one another, leaving no noticeable space or segmentation-cavity between them. After they have increased to about 32 in number, the blastoccele and its opening to the exterior (fig. 4, bl.c.) become recognizable. The embryo the morula stage is somewhat oblong in shape and has a quite spacious blastocole, and the blastocolic pore (bl.c.) is distinct on the ventral side (fig. 5). However, this pore disappears at an advanced morula stage, and apparently the vitelline membrane also, nearly simultaneously with it. At any rate both have altogether passed out of sight at the next stage, that of the blastula. In fig. 23, which represents a median section of a voung mornla (the outline of which has undergone mutual compression by the crowding together of embryos), the pore (bl.c.) is cut through and appears as a slit-like passage between two of the bounding blastomeres.

In the blastula (fig. 25) the wall consists of cylindrical cells and encloses a tolerably wide blastoccelic cavity, which is now at its greatest development. In this stage, the bilateral symmetry of the future larva is already established. It has an oblong planoconvex form, the flattened face of which corresponds to the future ventral face; and its ends, one somewhat broader than the other, indicate respectively the future anterior, and posterior, ends. The cells of the wall are all cylindrical in form, as shown in fig. 25, those of the ventral side being slightly larger than those on

the convex dorsal side. The nuclei in all the blastodermal cells are always situated in a peripheral position.

Plasmic corpuscles.—A noteworthy fact with regard to the blastula is that in its older stage a certain number of small and non-nucleated plasmic spheres is almost constantly met with in the blastocelic cavity (fig. 25, pl.co.) These have been first described by Foettinger under the name "corpnscules mésodermiques." According to this author, these corpuscles are free nuclei imbedded in a common protoplasmic mass which is supposed to fill up the blastocele, each corpuscle bocoming a mesoblast cell, after appropriating to itself a certain portion of the surrounding protoplasm. This view of Foettinger, which certainly can not be accepted at the present day, was, I believe, partially due to the then defective technique. His method consisted in ponring dilute acetic acid over the living embryo, and this, as the author himself was well aware, is highly detrimental, in that it frequently breaks up the blastomeres into fragments. The corpuscles described by him from so early a stage as that with only 8 blastomeres must have been simply produced by fragmentation, the result of his drastic treatment. The common protoplasmic mass supposed to be present in the blastoccele, was probably nothing but a coagulum.

Again the mesodermzellen which Metschnikoff ('82) found in the blastocelic cavity of the blastula are certainly not true mesoblast cells but rather certain spheres similar to Foetheres," as was rightly pointed out by Caldwell. Recently E. Schultze ('97) published a short paper entitled "Veber die Mesodermbildung bei Phoronis," in which he writes as follows:—"Schon auf dem Stadium der rundlichen Blastula schen wir einige Mesodermzellen im Blastocel aufsitzten"

(l.e., p. 6). It seems to me that Schulze has fallen into the same mistake as Metschnikoff.

Lastly, Caldwell ('82) has entertained a view quite different from those of other writers. According to him, the bodies in question are not present as such in the blastoccele, but are in reality only the cut ends of blastoderm cells projecting into the cavity and as such of course have nothing to do with the true mesoblast.

In the *Phoronis* studied by me, the plasmic corpuseles are present only in the highly advanced blastula (fig. 25, pl.co.). They are usually round in shape and very much smaller in size than any of the blastoderm cells, but as to structure, they do not show any deviation from the latter, except in the important respect that they have no nucleus. Although I tried with them all the available nuclear stains, the presence of any chromatic substance in them could in no instance be detected. In an earlier stage such a free floating sphere has never been met with. Instead of it, some unusually elongate blastoderm cells (pl. co., fig. 24), such as were found by Caldwell, were discovered protruding their inner end into the blastocæle. The nucleus of these cells commonly lies in the periphery as in all other cells of the more ordinary shape. In my opinion, the proximal ends of the clongate cells break off from the main cell-body and fall into the blastoccele, where they undergo degeneration, breaking into ever smaller and smaller spheres. By examining serial sections of au advanced stage like that of fig. 25, it is easy to convince one's self that there exists no connection whatever between the spheres and the blastoderm cells. The spheres, or the plasmic corpuscles, are clearly distinct bodies and not mere ends of blastoderm cells cut off in the process of microtomizing as was supposed by 518 1. IKEDA:

Caldwell. Their small size and the total absence of nuclear substance make it easy to distinguish them from the true mesoblast cells.

The corpuscles are still frequently discovered in the blasto-codic cavity at the beginning of gastrulation, together with a few mesoblast cells. But in an advanced gastrula they have wholly disappeared, possibly having been absorbed by the blastoderm cells. I think the temporary co-existence of the plasmic corpuscles and of the true mesoblast cells in the blastocode of the gastrula, has led some previous authors to confound the two elements. As to the significance of the corpuscles, I can at present offer no opinion "unless they be merely an excess of supply of nourishment analogous to food yolk" as has been suggested by Caldwell ('82, l.c., p. 18).

## c. Castrulation and Mesoblast-Formation.

In this section, I shall first describe what I conceive to be the true history of gastrulation and mesoblast-formation, and then pass on to a discussion of the views of other writers. The two developmental processes are so intimately related to each other, that it seems best to treat them together.

First as to external changes. The bilateral symmetry of the plano-convex blastula becomes more clearly marked than before when the gastral invagination begins on the ventral or the flattened side. The initial depression occurs over the whole ventral wall, so that a saucer-shaped embryo is produced. At first it is so shallow as to be perceived with difficulty in the surface view. Soon it deepens, becoming deepest at a point somewhat nearer to the broader end than to the narrow end of the embryo. The

deepest portion may conveniently be called the central depression. Fig. 6 represents the ventral view of an embryo in which the invagination has become visible from the outside, the central depression being most deeply shaded in the figure. In a slightly more advanced stage, as the original wide depression grows deeper. the external opening is gradually drawn together and at a certain stage (fig. 7) becomes transformed into an almost triangular blastopore situated at a position slightly anterior to the centre, as was the case with the central depression. The anterior side of the triangular blastopore is somewhat rounded by curving uniformly outwards, while posteriorly the two other sides gradually approach each other so as to meet at a point which may be called the apex of the triangle. Leading backwards from this apex, there runs in the median line the so-called primitive groove. This latter and also the triangular shape of the blastpore are occasioned, in my opinion, simply by the blastopore, originally broadly open, becoming narrowed by the special activity in the lateral posterior parts of its posterior section. In other words, the cell-multiplication of the ectoblastic layer is carried on especially in the last mentioned parts, so that there the pressure, which is exercised by the ectoblast towards the invaginated laver, is more marked than in the anterior and lateral borders of the blastopore. As the result of the above phenomenon, the definitive blastpore is pushed further anteriorly, and consequently, the archenteric cavity deepens in the posterior direction, as shown in fig. 29. The above consideration is supported by the results of actual measurements of the size of the embryos concerned. In spite of the fact that the embryo has developed considerably the body-length does not show any significant increase, remaining all the while at about 0.12 mm. on an

average. This shows that the growth is lost in the curvature of the body.

When the growing larva reaches the stage represented in fig. 8, the blastpore assumes a narrow transversely directed, slit-like form. That portion of the larval body lying in front of the blastpore—which is the persistent larval mouth—protrudes more or less prominently forwards and ventrally, so as to acquire the form characteristic of the preoral lobe of Actinotrocha. In such an advanced gastrula, the primary gut-cavity is well established and can be plainly traced through the wall in the surface view. If the larvae of such an early stage of development be taken out of the embryonal mass and set free in water, they will swim about by means of the well developed cilia, which cover the whole external surface.

Fig. 9 represents a side view of a larva, in which the preoral lobe has grown to a very considerable size. The nephridial pit, which is an ectoblastic invagination just in front of the posterior end of the gut, is now distinctly visible from the outside. In short, the larva may be said to possess the inceptive characters of an Actinotrocha.

I will now proceed to describe the internal changes accompanying gastrulation. The earliest symptom of this process can be seen in sections before it can be detected from the surface. It consists at first in a peculiar disposition of those blastodermic cells which constitute the ventral portion of the blastula wall. This portion not only shows a shallow concavity, but also the cells composing it become, as figs. 26 a and 26 b show, irregularly arranged on account of mutual pressure, as a result of which some of the cells are even forced out of file so as to fall into the blastocode. These liberated cells have usually a round shape and

of course contain each a distinct nucleus. Some other cells are apparently in the process of being pushed out and have a club-like shape, the narrowed end being still inserted between the cells of the layer to be invaginated. A further symptom of incipient invagination consists in the circumstance that the nucleus in most cells of this portion has no longer a peripheral position, but is situated in the middle or rather nearer to the inner end of the cells. Moreover, the nucleus is frequently met with in the form of the karyokinetic figure which shows that the cells are dividing and increasing in number in the layer to be invaginated. The cells pushed out into the blastocale are nothing clsc than mesoblast cells, so that it may be stated that the mesoblast-formation begins simultaneously with the gastrulation.

At the beginning of gastrulation we can thus distinguish two parts in the blastoderm wall, viz., the mesentoblast and the ectoblast. The former corresponds to the whole of the portion to be invaginated, while the ectoblast forms all the remaining portion of the embryo. The mesentoblast is composed of large and irregularly arranged cells, while the ectoblast is of taller cylindrical cells regularly arranged in a single row (figs. 26 a and 26 b). The mesentoblast, as the name indicates, is destined to give rise to both the entoblastic and mesoblastic elements. The characteristic disposition of its constituent cells indicates its being the source of mesoblast proliferation.

The mesoblast proliferation becomes more and more accentuated in activity as the gastral invagination gradually deepens (see fig. 27), but the mesoblast cells thus formed do not adhere as a lining epithelium to the ectoblast, until what are called the anterior diverticula have been formed on both sides of the blastopore. The blastocole, in which the mesoblast cells are at first loosely

scattered about, is henceforth greatly reduced in extent and finally, as the development of the archenteron progresses, is almost obliterated, especially along the dorsal and lateral portions of the embryo where the ectoblast and the gut come into direct contact with each other (see figs. 29 and 30 b).

Figs. 28 a-c show three cross sections through different parts of a larva of nearly the same stage as that represented in fig. 6, in which the invagination has become recognizable in the surface Fig. 28 a passes through the central depression which becomes gradually shallower posteriorly (figs. 28 b and 28 c). these figures show, the mesoblast cells are at this stage still being proliferated uniformly from every part of the mesentoblast and do not vet form a lining epithelium to the ectoblast. When the blastopore has taken a triangular shape (fig. 7) and the primary archenteric cavity has somewhat bent itself towards the hind end, the posterior border of the blastopore has travelled a certain distance in an anterior direction. If we examine serial sections of this region, a narrow and shallow groove is detected running for a short distance immediately behind, and from, the meeting point of the lateral blastopore lips. Also at about this stage, a paired invagination, the anterior diverticula of Caldwell, appears along the side of the anterior portion of the archenteron. These points will become clear from a consideration of figs. 30 a-c, which are drawn from serial transverse sections of an embryo slightly older than that shown in fig. 7. In fig. 30 a, showing the right-hand side of the blastopore, we notice a lateral infolding (ant. dir.) of the archenteric wall a short distance inside the blastopore lip. Here the component cells are irregularly arranged and their entire disposition reminds one of the mesentoblastic layer. Indeed some indubitable mesoblast cells are found pressed

against the tip of the diverticulum. No doubt the mesoblast is here arising, not by direct cell multiplication, but by the pushing in of the cells of the diverticulum. This is more clearly illustrated in fig. 31, which shows a transverse section through the blastopore of a more advanced larva; here the mesoblast cells almost fill up the blastoccelic cavity on both sides of the blastopore. In fig. 30 b, a transverse section just behind the closure of the blastopore, the most anterior portion of the primitive groove before mentioned is cut across. Here the wall of the groove underlying the gut is formed of mutually compressed cells, some of which are evidently migrating into the blastocele (on the left-hand side in the figure). If the sections are followed further posteriorly, the groove still persists, but no mesoblast cell in the actual immigrating process can be discovered, although there are those which have been previously pushed out and are now floating between the two primary germinal layers at this region. Still more posteriorly the groove entirely disappears and the entoblastic and ectoblastic layers are separated from each other by the comparatively wide blastoccelic cavity (fig. 30 c). At this stage, therefore, the greater part of the archenteric wall has ceased to contribute towards the mesoblast-formation; in other words, it has lost its mesentoblastic nature. The mesoblast is now being produced only from two limited regions, viz., autorior diverticula and the ventral groove.

In a slightly more advanced larva, the ventral groove is still present for some distance immediately behind the blastopore, but the layer which forms the groove has entirely ceased to give rise to mesoblast cells (fig. 32, which is taken from a transverse section very near the blastopore). It appears to me that this groove is to be regarded as but the posterior portion of the original mesentoblast, which, owing to the fact that the central depression

is eccentrically placed nearer to the anterior end, has to traverse a longer distance before it can be reflected inwards, and thus on its inward course lags behind the anterior and antero-lateral portions. Eventually all the cells of the wall of the groove that are left behind after proliferating the mesoblast cells, are without doubt invaginated and form a part of the entoblast. The groove then entirely disappears. I could not discover any remnant of it in any part of the posterior region where, according to Caldwell, the ectoblast and the entoblast are said to stand in fusion to give rise afterwards to the anus. In such an advanced stage, the anterior diverticula have also ceased to give off mesoblast cells and have become straightened out, their walls acquiring a normal epithelial character (entoblastic).

From the facts above adduced, it may be concluded that both the anterior directicula and the ventral groove, present at a certain developmental stage of the Phoronis embryos, are remnants of the original mesentoblast which at an earlier stage occupied the the whole extent of the gastral invagination. They are, therefore, merely temporary, and destined sooner or later to split into mesoblastic and entoblastic cells.

As will be seen in figs. 30 a-c, the ectoblast and the archenteric walls are brought together into such close contact, especially along the dorsal and lateral regions that scarcely any interspace is left between them. In the embryo given in fig. 8, the cavity of the rudimentary preoral lobe is filled with mesoblast cells produced from the original mesentoblastic layer. So far as I can make out, these show no difference whatever from those proliferated from the anterior diverticula; both are indistinguishably mixed together. Though most of the mesoblast cells in the preoral lobe lie loose during the active period of the diverticula, there are

found a few that have already apposed themselves flatty to the ectoblast (see fig. 29), while the eavity behind the blastopore still remains without a mesoblastic lining. This last condition persists till the period when the nephridial invagination makes its appearance. The state of things in question is to be seen in fig. 29, which represents a median sagittal section through an embryo of nearly the same stage as fig. 8.

Soon afterwards the anterior diverticula and the ventral groove entirely disappear and the preoral lobe begins to bend more distinctly downwards. Meanwhile an unpaired ectoblastic invagination appears at the posterior end of the larva, on the ventral side of the blind end of the now greatly elongated gut. It appears at first as a shallow depression (fig. 33, nep. p.) of purely ectoblastic nature, having nothing to do with the mesentoblast. It is from this invagination that the future nephridia of Actinotrocha develop and hence I shall call it the nephridial pit, in preference to the name "anal pit" of CALDWELL, who for the first time described this structure. I have very frequently noticed signs of vigorous cell-division in the cells of the pit wall, evidently only for enlargement of the pit itself, since the axis of the karvokinetic spindle is always placed paratangentially to the wall. I have moreover often noticed peculiar ectoblastic cells round in shape and in process of multiplication, situated just outside the edge of the entrance to the pit (fig. 33).

In larvæ of the stage of fig. 9 the nephridial pit can be well seen in surface views. This stage further attracts our special attention on account of several important developmental processes taking place in it. First to be noted is the fact that from the posterior end of the primary gut a small and short evagination protrudes itself touching the ectoblast with its blind posterior

end. This hollow protuberance is the rudiment of the intestinal canal of Actinotrocha. In longitudinal section it is shown in fig. 37 (int.).

In fig. 34, representing a slightly oblique frontal section of a larva of nearly the same stage as that of fig. 9, we see below the pit-like nephridial sac, which is quite free from the gut. The ectoblastic wall of the preoral lobe is at this stage somewhat uniformly lined with flattened mesoblast cells, while in the cavity behind the blastopore the mesoblast cells are for the most part freely scattered, though a few have already begun to arrange themselves against the ectoblast layer in this region. In fig. 35, a transverse section through the posterior end of a larva of nearly the same stage, the nephridial pit appears as a single flattened sac (nep. p.) lying in front of the intestine (int.); the ectoblastic wall is internally lined with a few isolated and flattened mesoblast cells. In a slightly more advanced stage, the ectoblast behind the blastopore, and in a less complete degree the gut wall also, shows a similar mesoblastic lining, though a few mesoblast cells still remain free, especially in front of the nephridial sac.

In order to facilitate comparison with the statements of other writers, I will here add a few words on the change of form undergone by the nephridial pit. When in a larva slightly older than that of fig. 9, the preoral lobe and the future intestinal portion of the gut have become considerably elongated, the nephridial pit, which has meanwhile become deeper than before, begins at its inner blind end to divide into two lateral branches. Each of the latter corresponds, as will be fully demonstrated further on, to the nephridial canal of Actinotrocha. Fig. 38, a frontal section of a larva at this stage, shows the bifurcation just alluded to. The

relation of the unpaired nephridial sac to the gut will be best understood from the median sagittal section given in fig. 37.

I may here be allowed to put in a short historical review of the mesoblast-formation in the *Phoronis* larva.

Kowalewsky ('67) attributed the origin of the mesoblast to the ectoblast.

METSCHNIKOFF ('82), FOETTINGER ('82), and E. SCHULTZE ('97) confounded the plasmic corpuscles with the true mesoblast, and none of them was aware of the presence of the anterior diverticula.

Caldwell ('85) made many interesting observations on the mesoblast-formation. According to his view, there exists no mesoblast before the closure of the blastopore lips (lateral), but it arises later from three distinct sources, viz., 1) the anterior paired diverticulum (entoblastic), 2) the posterior paired diverticulum (ectoblastic) and 3) "the primitive streak" connecting the above two structures. Further it has been declared by him that the body-cavities of the larva arise in two different regions. As to the preoral body-cavity, he writes as follows: "From the time when two or three mesoblast cells are budded off from the directicula on either side, a cavity is present in each mass thus formed. These eavities are the two halves of the body-cavity (preoral)" (l.c., p. 374). On the other hand with respect to the posterior body-cavity, he states that "it is formed independently in a paired mass of cells which grow out to the end of the first formed sacs and remain separated from septum" (l.e., p. 376). Thus he regards the preoral body-cavity as arising after the enterocelic type. Lastly the author puts forward in his recapitulation the opinion that the blastopore gives rise to both the mouth and the anus.

Roule ('90) also distinguished two sorts of mesoblast cells in view of their different origin and fate: "Mesenchymes primaires" and "initales mésodermiques." Both are derived from the "protoendoderme" which forms the primary archenteric wall. The latter gives rise to cells grouped together into two compact "bandlettes mésodermiques." which are regarded as homologous with the mesodermal bands of Annelid larvae. In reality these bands are, as have been pointed out by Schultze, nothing else than the posterior paired diverticula of Caldwell.

From the account of the mesoblast-formation given in the foregoing lines, it is evident that the first stages of that process are observable from the very beginning of gastrulation (figs. 26 a & b), and long before the blastopore takes the small triangular shape. On this point my observations stand at variance with Caldwell's. Nor can I agree with that author in the opinion that the mesoblast produced from the anterior diverticula (even though consisting of only two or three cells) incloses an enterocedic cavity. As already described, the cells in question, after being budded off, lie loose in the blastocede together with preexisting mesoblast cells and without forming a wall to a special cavity of any sort.

As to the ventral groove, Metschnikoff ('82) was the first to refer to this structure and wrote as follows: "In passender Lage des Embryos kann man eine in Verbindung mit dem Blastoporus befindlichen Furche (longitudinale) wahrnehmen, welche zum Hinterende des Embryos hinzieht und sich nur auf dem Ektoderm beshränkt. Diese Furche erhöht den bilateralen Bauplan des Embryos erscheint indessen als eine vergängliche Bildung, welche man auf späteren Stadium vergebens suchen würde" (l.e., p. 301). According to Caldwell, this groove, which he calls "the primi-

tive streak," is produced by a fusion of the blastopore lips; the cells along the fusion line differentiate after multiplication into the epiblast, the hypoblast, and the mesoblast. And the rapid growth of the epiblast in this region soon obliterates the groove, leaving however its posteriormost portion as the "anal pit." But such, as I have tried to show, is not the case, for the so-called primitive streak entirely disappears leaving no trace whatever, long before the nephridial or anal pit makes its appearance. Therefore there exists no direct genetic relation between the primitive streak and the anal pit.

Caldwell's view that the two nephridial ponches give off the mesoblast, which eventually lines the posterior body-cavity, can not be sustained; for, according to my own observations, that body-cavity with its mesoblastic lining wall is already present before the nephridial pit divides into the two pouches. It is true that the cells floating in the posterior body-cavity are in some sections found aggregated at the blind ends of the ponches as shown in Fig. 38. This is a condition which might mislead one to the conclusion that mesoblast cells are here in process of proliferation. But solid cell accumulation in such a section is to be considered as simply due to the obliteration by compression of the lumen of the nephridial pouches. Fig. 36 taken from an obliquely cut sagittal section through a larva of this stage, shows no wandering cells in front of the pouches (of which only the right one is seen in the figure); in this case there is certainly no doubt about the matter.

Finally as to the anus, Caldwell mentions "a solid cord of cells" which he considers to be the posterior remnant of the primitive streak. According to him, this acquires a lumen and forms a fine canal leading from the primary gut cavity to the

exterior. However, it seems clear to me that this cord is nothing else than an early stage of the intestinal outgrowth independently produced at the posterior end of the gut. Moreover, in *Phoronis ijimai*, the gut cavity does not come into communication with the exterior at so early a developmental stage as Caldwell observed; in that species, the anus first opens at a definite stage when the larva bears two pairs of larval tentacles.

E. Schultze ('97) rejects Caldwell's views in regard to the anal pit, but regards it as a rudiment of the future ventral pouch of Actinotrocha. This is, however, certainly not true, since the ventral pouch is a thing that has a distinct origin and appears at a much later stage of larval development.

# d. Further Observations on the Development of the Larva.

Some authors have recorded that the larva swims about abroad at such a stage of development as is represented in fig. 8. However in *Phoronis ijimai*, the larva lies hidden in the lophophoral loops of the mother until it has acquired at least two pairs of larval tentacles.

In the larva shown in fig. 9, the somewhat prominent preoral lobe hangs over the larval month. Local ectoblastic thickenings occur at two places, viz., at the centre of the upper surface of the preoral lobe and along the mid-ventral line near the posterior end of the body. The former is the future nerve ganglion; the latter, the rudiment of the first pair of larval tentacles. The nephridial invagination at the posterior end is still shallow.

At a little later stage, the tentacular thickening divides into

two more prominent ridges running on each side obliquely anteriorly. The preoral lobe grows rapidly so as to hang down on the ventral side and as a consequence of this an esophageal canal is formed (fig. 37,  $\omega_8$ ). The asophageal wall is, therefore, ectoblastic in origin and is composed of strongly ciliated columnar About this period the nephridial invagination becomes completely divided into two lobes at the proximal end, as I have already described (figs. 37 and 38, nep. p.). In more advanced larvæ, the pit is split throughout its entire length into two nearly parallel canals, each of which opens independently to the exterior. Figs. 39 a-c show three transverse, though not consecutive, sections passing through the posterior region of a larva at such a stage. In the first of these figures, the two cell-masses (nep. c.) on either side of the stomach represent the uppermost portion of the nephridial canals. In the second figure, each of the cell-masses encloses an easily distinguishable lumen. The two canals finally open to the exterior each by a small pore (nep, o), as seen in the third figure (only one pore is cut through in the above figure, the section being slightly oblique to the main axis of the larval body). In the above figure we see an ectoblastic cell-mass separating the right and the left nephridial canals (nep. c). How is this partition brought about? I think it is caused by reevagination of the distal unpaired portion of the nephridial pit, as by that process the pit wall forming the above portion is gradually transferred to the body-surface of the larva.

Meanwhile the esophagus becomes more and more elongated, while the paired tentacular thickenings bulge out each into two perceptible prominences. The latter represent the rudimentary state of two larval tentacles, each of which has internally a cavity continuous with the postoral body-cavity. Fig. 10 represents a larva

with two pairs of as yet very short larval tentacles; this is the most advanced developmental stage to be met with in the embryonal masses. Fig. 40 is a median sagittal section of such a highly advanced larva. Here the osophagus (os.) and the intestine (int.), which latter now communicates with the exterior by the small anus (on.), are highly developed, so that the three parts of the alimentary tract (osophagus, stomach, and intestine), may be said to be almost complete. The nerve ganglion (fig. 40, gl.) is well differentiated from the cetoblast of the preoral lobe, presenting itself in section as a round, well marked mass principally composed of nerve fibres. I have been unable to ascertain whether a proctodeum is produced at all, and if so, what part of the postgut it gives rise to.

The preoral body-cavity is, at this stage of development, still very incompletely separated from the postoral cavity by a few mesoblast cells (fig. 43, mes.). The nephridial canals (fig. 41, nep. c.) are now distinctly separated and removed from each other, and are found in a cross section to be situated laterally to the intestine (int.). One on the right-hand side of the above figure is cut through at its external opening, while on the other side the nephridium is represented by a thick mass of a few ectoblastic cells. This lateral shifting of the nephridia becomes more and more pronounced with the advancement of larval development. A slightly advanced state of the nephridia is shown in fig. 42, where the nephridial canals (nep, e) are now seen tolerably long and have a wall composed of a single row of cubical cells. It is often observed that some mesoblast cells connect the canals with the splanchnic walls (see the above figure). These cells seem to be the first indication of the future collar-trunk septum. Besides, a certain number of mesenchymatous cells, which later undoubtedly become the excretory cells of Actinotrocha, is always found attached to the blind ends of the nephridial canals. Caldwell says that he saw the excretory cells aggregated around the apex of each canal and that they had numerous plasmic processes, giving them a strong resemblance to the perforated cells known in *Echiurus*. It seems, however, highly probable to me that this strange appearance of the excretory cells is an artefact, since, as I shall point out later, the same cells in Actinotrocha are certainly not provided with any such processes.

I have very frequently detected some gigantic mesoblast cells floating freely in the postoral body-cavity of larvæ with one or two pairs of tentacles (fig. 44, corp.). They are round and nucleated and contain numerous large yolk-spheres. After repeated examination I have come to regard them as mother-cells of blood corpuscles which are found as corpuscle-massess in the collar cavity of Actinotrocha. This point will again be treated of in detail in the proper place in the following section.

#### II. The Structure of Actinotrocha.

#### a. External Appearance.

It can scarcely be doubted that each species of the *Phoro-nide* has a characteristic form of Actinotrocha peculiar to it. Some of the previous observers (e. g., Wilson and Masterman) have mentioned two distinct types of larvae as occurring in the same locality. Among the larvae which I observed at Misaki, I was able to distinguish four different types, each of which had a characteristic form and a more or less definite topographical

distribution. I will designate these types by the letters A, B, C, and D.

Type A (fig. 13). The larvae of this type were principally collected in Aburatsubo and belong in all probability to the species *Phoronis ijimai*, which, as I have said, is found in the same locality. The body is comparatively short and thick, measuring about 1.–1.5 mm, in total length. The larval tentacles of a full grown larva never exceed 16 in number.

Type B (fig. 14). This is a larger form than the preceding (about  $2-2.5 \, mm$ , in length). The body and the intestinal canal are long and slender. The full grown larva has about 28 tentacles which are much more slender than those of Type A. Peculiar to it is the sensory spot (so.) situated just in front of the ganglion (gl.). The larvae were found in greatest abundance near Kitsunesaki, a point at the mouth of the inlet Moroiso.

Type C. (figs. 15 a & b). This type is distinguished from all the others by several characteristic points. In size of body it is intermediate between Types A and B (usually 1.5 mm, in length). The body is relatively short and thick. The number of tentacles, so far as I know, ranges from 16 to 24. A pair of flask-shaped glands (gld.) is found one on either side of the ganglion (gl.). A pair of retractor muscles (ret) runs longitudinally through the trunk cavity from the tentacular ring to the apex of the anal cone. Compared with the first two types this is much rarer.

Type D (figs. 12 and 16). This is a rare form of which I have obtained only seven specimens in all. It is enormously large in comparison with the others (4.–5, mm, in total length and 1, mm, in width). The preoral lobe is disproportionately small, while the trunk is long and thick. The tentacles are remarkably

numerous, sometimes reaching 48 in number. In a single living specimen, the skin of the trunk was of a light orange colour; the subdermal circular muscles were especially well developed in the trunk but interrupted at four longitudinal clearly marked zones.

The youngest swimming larva I have ever obtained was of type A. It was already supplied with four pairs of tentacles which, however, were still short. The body measured about 0.5 mm. in length. The trunk was short and showed a slight characteristic curvature, the concavity being turned toward the dorsal side. The thickly ciliated hood was comparatively large; the ganglion and the perianal ciliated belt were already well developed. In the surface view of this larva during life, I was not able to detect the ventral pouch nor the corpuscle-masses.

At about the stage with five pairs of tentacles, the trunk becomes clongated and straightend out. The nephridia may then be seen in their characteristic bouquet-form, and the ventral pouch appears as a solid ectoblastic thickening. Neither the corpuscle-masses nor the retractor muscles are yet to be seen.

As the larva grows, the number of tentacles increases in pairs proceeding from the ventral side toward the dorsal; hence, the most dorsally situated tentacles are the youngest and the shortest. In larvae with 12 tentacles and belonging to type A, the ventral pouch is deep enough to be plainly visible from outside. We always notice from this stage on a pair of the retractor muscles which extends between the ganglion (yl.) and the dorsal inner side of the tentacular circle (ret. in figs. 12, 13, 14, and 15).

The larval organisation of types A and B is nearly completed in the stages with 14–16 tentacles. Let me next give a somewhat detailed description of the external appearance of Actinotrocha in general.

536 1. IKEDA:

The preoral lobe. This is a structure which looks like a broad hood with its concavity directed downwards. It almost entirely covers the upper anterior part of the collar,\* when not influenced by external circumstances. Masterman has made the statement that in its natural attitude the hood has its length disposed parallel to the principal body-axis. However, if the larva be examined in the living state, it will at once be discovered that its normal disposition is horizontal. It becomes turned up only as the result of preservation. Its whole surface is covered with cilia, most strongly developed along the free margin which constitutes the preoral ciliated belt. In the full grown larva, the ganglion (and also the sensory spot in type B) has also a set of specially long cilia on the outside. Numerous fine and refractive nerve fibres are seen radiating from the ganglion (gl.) to the free margin of the lobe (pre. bel.) (figs. 13 and 14).

Masterman has described and figured two ectoblastic structures which are said to be situated on the ventral wall of the hood and which he has named the "oral" and the "pharyngeal" grooves. These he compares, as to their function, to the gill-slits of the Chordata. I can not but think that that writer has here fallen into a very grave error, which might have been avoided, had he examined the structures in question in living specimens. Among the preserved specimens I have frequently noticed those in which the lower or oral wall of the hood was prominently bulged out in front of the month. In consequence of that prominence (fig. 16, prom.), there was produced on either

<sup>\*1</sup> adopt this name of Masterman's to denote that portion of the larval body which lies in front of the tentacular circle and behind the preoral lobe.

side of the mouth a transverse groove, which was visible when viewed from the ventral side. I believe it was to grooves of this kind that Masterman assigned the above important significance. In my opinion, they are simply artificial productions due to preservation.

The Collar. The form of the collar as a whole may be compared to a cylinder obliquely truncated at the posterior end. Its posterior border is fringed with a regular row of tentacles, while anteriorly it is joined to the hood by a narrow neck. The number of tentacles (larval) varies according to the different stages of growth and also according to the type to which larvæ belong. They are most numerous in type D, most individuals of which bear 40-48 tentacles (figs. 12 and 16). The rudiments of the adult tentacles make their appearance as bud-like ectoblastic thickenings immediately below the base of the larval tentacles. An exception to this rule is found in the case of larvæ belonging to type D, in which the adult tentacles are represented by a local ventral thickening of the wall of the larval tentacles at their proximal portion (see fig. 58 d, s. t.). It is very probable that the number of the larval tentacles corresponds to that of the adult. In type A, at any rate, I have ascertained that the full grown larva and the worm just metamorphosed bear the same number of tentacles, namely 16.

The trunk. This portion, which is the shortest of the three regions in early larval stages (fig. 10), comes with growth to occupy the largest part of the larval body and assumes a long cylindrical form. Its anterior boundary is the tentacular circle; the posterior end is girdled with the perianal ciliated belt which serves as the larval locomotory organ.

### b. The Internal Structure of Actinotrocha.

## 1. Body-Divisions and Body-Cavities.

I have endeavored to show in the preceding pages, that the body-cavities of Actinotrocha do not arise from the enteric diverticula, as was insisted upon by Caldwell, but that they are simply produced by mesoblastic cells applying themselves to and forming the lining of the ectoblastic, and the entoblastic, wall. They may, therefore, be classed under the "pseudocole" or "schizocale" of Herrwig. Moreover, the body-cavities of Actinotrocha as a whole do not in their genetic relation correspond to those of the adult, as I shall attempt to elucidate in the sequel. During the metamorphosis, the greater part of the former (the preoral cavity) is almost wholly lost, while the other part (the collar-cavity) is transformed into a vascular space, so that what is known by the same name in the adult is of an entirely new origin. Thus we see that the larval body-cavity of Actinotrocha, i. e. the trunk cavity, is the only portion that persists among the bodycavities of the adult, in which it is known as the foot or infraseptal cavity. In correlation with this circumstance are observable certain changes in the position of the nephridia and of the vascular As described by Caldwell, the nephridia of Actinotrocha, which are not provided with an internal opening, lie for the most part in the collar cavity, while after the metamorphosis they are found wholly in the infraseptal cavity of the worm. the paired corpuscle masses which are found only in the collar cavity of the larva, are no longer seen in the same cavity of the adult. These changes to a certain extent at least establish the

fact that some profound changes in the arrangement of the body-cavities must occur during the metamorphosis. As is acknowledged by all, the supraseptal cavity of *Phoronis* is greatly reduced in size as compared with that of the larva, and contains almost no organ except the blood vessels. The infraseptal cavity is, on the contrary, very wide, and contains many important organs, e. g., the alimentary canal, the sexual organs, and the main part of the vascular system. Thus it becomes necessary to make distinctions between the body-cavities of Actinotrocha and those of the adult and to call them respectively by different names. The former may be termed the larval body-cavities, and the latter, the adult body-cavities.

Most previous writers have not taken any particular notice of the relation which exists between the external body-divisions and the body-cavities of Actinotrocha, so the words "hood" and "foot" do not denote anything but mere external features. The idea of segment was first introduced by Caldwell; he considers the larval body as divided into three parts: (1) the preoral lobe set in front of the septum, (2) the trunk portion situated behind the septum, and (3) the foot or invaginated pouch. According to this view, the body-cavity is divided by the septum into two contiguous parts, viz., the preoral cavity in front of, and the trunk cavity behind, Masterman divides the entire body into three the septum. portions, viz., the preoral lobe, the collar, and the trunk. These three divisions are not only externally marked by their respective forms, but also by the presence of two transverse septa or mesenteries. Thus we see, the preoral lobe of Caldwell comprises both the preoral lobe and the collar of Masterman.

Whatever may be the value of Masterman's Diplochorda hypothesis, I feel inclined to accept with some modifications, his view of the body-divisions. The external appearance of the three portions I have already described in brief. As to the internal body-cavities corresponding to these external portions, I can not agree with Masterman, when he says that they are completely separated from one another; for, as I shall soon show, the septum which lies between the preoral and the collar cavities is always an incomplete formation, at least in all the Actinotrochæ which I have observed. Besides, I have been unable to detect the first and third pairs of nephridia, which are said to exist in the preoral, and the trunk cavities (Masterman). Therefore, I can not regard the body-divisions of Actinotrocha as "segments" in the sense of that author.

The septa or mesenteries are very delicate in structure and can hardly be recognized in living specimens. I have, therefore, had to study them mostly in sections. I shall hereafter call the two septa the preoral, and the postoral, septa.

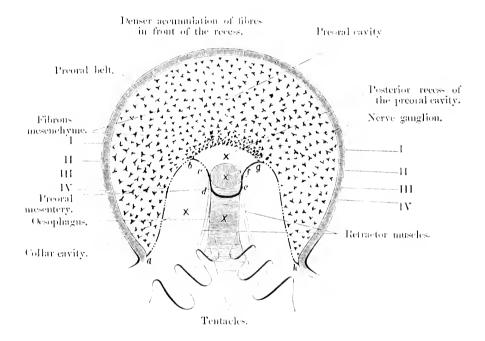
Larval Preoral Body-Cavity. The larval preoral body-cavity fills up the interior of the hood, in which there is no entoblastic organ. Innumerable mesenchymatous fibres traverse the cavity (figs. 45, 49, 63 a, m.f.). A few blood corpuseles are also frequently discovered in this cavity (fig. 49, corp.); this fact is, I believe, one of the proofs of the correctness of the view which I now propose to consider.

I have already spoken of the incomplete formation of the preoral septum. Whether this is a mere specific difference or not, remains to me uncertain, as I have had no chance of examining the larva investigated by Masterman. In sagittal sections of the larva at any stage of its growth, the septum can constantly be traced so long as the cosphagus is contained in the sections. In figs. 45 and 63 a, a slender cellular strand (mes'.) behind the ganglion (gl.) represents the septum in cross section. It extends between the

upper and the lower walls of the hood. Thus it will be seen that the septum completely separates the preoral cavity from the collar eavity just behind the ganglion. But, when we come to sections passing through a more lateral region to either side of the ganglion or of the esophagus, the upper portion of the septum becomes abruptly indistinct. In fig. 54, which shows a sagittal section through the right-hand side of the œsophagus of a larva of 16 tentacles (type A), the septum (mes'.) near its ventral attachment is indicated by a comparatively thick layer of cells, while the dorsal portion is divided into fine protoplasmic branches, of which some extend to the upper wall of the hood and others stop short As the relations of this septum are somewhat complicated, I will try to make them clear by referring to a series of cross sections (not continuous) through the hood of a larva of type D(figs. 59 u-d) and also to the annexed wood-cut. The latter is a diagrammatic representation of the Actinotrocha hood and its neighbouring part, as seen from above, i. c., in horizontal projection. The dorsal side is above and the ventral, below. Nearly in the centre is the nerve ganglion. Below it and concealed from sight, is the mouth, from which the esophagus leads downwards. The little stellate markings, scattered over the greater part of the figure, are supposed to represent mesenchymatous cells, which, with the branched and reticulate fibres arising from them, pervade the preoral body-cavity, except in a small space immediately in front of, and below, the ganglion. This free space I shall call the posterior recess of the preoral cavity. The line a b c d e f g h, curved somewhat like the letter M, indicates the position of the preoral septum. The part shown in full line represents that portion of the septum which is complete in structure and the part in broken line, that portion of the same which is incomplete. All

512 1. ikeda:

the space in front of the septum is the preoral cavity, while back of it lies the collar cavity.



If we now study the series of sections (figs.  $59 a - d^{\circ}$ ), the nature of this septum will become clear. Section 59 a is the most anterior of the four and passes through the hood at about the plane of the line I-1 in the above wood-cut. The whole of the preoral cavity is filled with the fibres of branching mesenchymatous cells except in the ventral median part (p.r.). This is the beginning of the posterior recess of the preoral cavity, which is limited anteriorly by a faint membraneous layer consisting of protoplasmic fibres only. Section 59 b is from about the plane of the line H-II. This contains the ganglion (gl.) and on each side

<sup>\*</sup> Unfortunately in the series of sections tissues have undergone considerable disturbance by the action of the killing reagent, but the relations of the layers remain unaltered.

the anterior end of the collar cavity (col.c.). Below the ganglion, the posterior recess (p.r.) is seen to have a complete wall, that is to say, the posterior septum is fully developed. In the above figure, the two wide spaces lying on both sides of the posterior recess correspond to the two anterior horns of the collar cavity projecting forward (marked b and g in the wood-cut). And we can certainly see that on the dorsal side as well as laterally there is no distinct partition or continuation of the preoral septum which, according to Masterman, should entirely divide the preoral and the collar cavities at every point. As the figure shows, the dorsal portion of the mesentery (mes') is decomposed into fine protoplasmic processes which join with those of the fibrous mesenchymes dispersed through the preoral cavity. In section, 59 c, passing though the middle of the ganglion (the line III-III of the wood-cut), the collar cavities (col.c.) are much wider and have become united below the esophagus (es). The septum (as the wall of the posterior recess, p.r.) in this region is a little more definite in form than in the last figure; the posterior recess (p.r.)is distinct as before. In section 59 d passing through the line IV-IV of the wood-cut, the posterior wall of the recess (p.r.) is obliquely cut and appears in the right-hand lower corner as a membraneous slice, the recess being distinctly bounded by the septum (mes'.). Outside of it are seen, one on each side, the sections of the retractor muscles (ret.), of which more will be said later. The collar cavity (col.c.) is now very spacious, but the septum laterally remains in the same condition as before.

From the above descriptions, it will be clear that the preoral septum is complete only in the median portion (indicated by the full line *c d e f* in the wood-cut), while in the more lateral part on each side, it is at the best a loose open reticular membrane,

544 1. IKEDA:

through which the colomic fluid of the preoral and the collar cavities is put in free circulation.

A questionable structure has been described from the preoral cavity by Masterman under the name "subneural sinus," and is compared to the structure bearing the same name in the Hemichorda. According to him, the subneural sinus is an interstitial space left between the two lamina composing the preoral septum, just under the ganglion and above the so-called "subneural glaud." Anteriorly and laterally, it is said to be surrounded by the preoral cavity, and posteriorly, by the collar-cavity; its upper and lower walls are claimed to be directly formed of the ectoblast without a peritoneal layer. Further it is said, that the sinus communicates mid-dorsally with the dorsal blood vessel on the esophagus. After repeated examinations of the larvæ of the four different types, I am convinced that Masterman's subneural sinus is identical with what I have called the posterior recess of the preoral eavity. It has nothing to do with the tissue-space in the preoral septum, but is clearly a part of the preoral body-cavity, which is free from the meseuchymatous fibres. Besides, I can not in any way detect the presence of the dorsal vessel on the œsophagus, a vessel which connects the subneural sinus with the dorsal vessel on the stomach. A view similar to mine as above expressed was given by Harmer in his paper on Cephalodisus ('97).

Masterman has further given an interesting description of the "probose pores," situated on each side of the ganglion. They are compared to the probose pores of Balanoglossus and are said to fulfill the same function as the collar nephridium of Actinotrocha. In the larvae studied by me, the only things that bear even a remote resemblance to them, are the flask-shaped glands which are seen on the upper face of the preoral lobe of the larva

belonging to type C. But the position of these glands in relation to the ganglion as well as their histological structure at once reveal their true nature. The internal openings of the organs were described by Masterman as follows: "Just where the preoral mesoblastic wall slopes away on either side of the sinus there are a pair of thickenings, which traced forwards, show themselves to be the commencement of a pair of internal openings" (l.e., p. 307). The paired thickenings referred to by him are apparently nothing else than the points of attachment of the retractor muscles in the collar cavity, as will be seen in fig. 59 d (ret.). Further details respecting these muscles will be given later.

Larral Collar Cavity. The collar-cavity is a comparatively wide space extending between the preoral and the postoral septa. It is produced anteriorly into two horns, embracing between them the posterior recess of the preoral cavity. It is perfectly separated by the postoral septum from the trunk eavity. The postoral septum, or simply the septum, as it is more commonly called, is stretched obliquely transversely between the splanchuic and the somatic walls, along a line a little below the tentacular circle (figs. 45, 48, mes.). Its dorsal attachment on the splanchnic layer is, as represented in fig. 45 (mes.), found at the plane of the junction of the æsophagus with the stomach, while ventrally the attachment lies much further below. In frontal sections of the larva, the septum (fig. 48 mes.) is seen on either side of the stomach and its somatic insertion lies just under the tentacles, so that each tentacular cavity is continuous with the larval collar cavity (fig. 45).

The adult collar eavity, or the supraseptal cavity, is already formed in the fully developed larva of every type, as a ring-space running along the inner side of the tentacular circle and above

the septum (see figs. 58 a and d, s.c.c.). This, together with several other larval organs in the larval collar cavity, had better be treated at a more suitable place in the sequel.

Masterman has described a dorsal mesentery running along the mid-dorsal line of the esophagus, and separating dorsally the larval collar cavity into two lateral halves. In the Actinotrochae of all the types observed by me and at every stage of the larval growth, no such mesentery is present. It is true that the body walls and the esophageal walls very frequently come close together, especially in the young larva after preservation, so as to greatly narrow the collar cavity in this region (figs. 49 and 50 a). But a mesentery is never to be found. Its absence is quite clear in the large Actinotrocha belonging to type D, in which the skin and the esophagus lie well separated by a considerable space (figs. 58 a and 58 b).

Trunk cavity. The trunk cavity occupies the interior of the third body-division—the trunk. It is completely separated by the postoral septum from the collar cavity, and since the septum is oblique in position, it extends dorsally nearly to the base of the esophagus. The ventral mesentery extends along the median ventral line of the body wall and of the alimentary canal, and is wholly confined to the trunk cavity. In fig. 45, which shows a median sagittal section of a young larva of type 1, a portion of this mesentery (r.mes.) is represented as a thin cellular membrane extending between the alimentary canal and the ventral pouch (p.o.), the latter being still shallow at this stage. The whole extent of the ventral pouch is stretched by the ventral mesentery to the skin as well as to the digestive canal. This relation remains the same as the pouch grows in length and finally winds around the digestive canal. A transverse section through the

trunk of a highly advanced larva of type C, is given in fig. 57 a, in which the much elongated and convoluted pouch is seen cut into several sections (po.), connected with one another by the mesentery (v.mes.).

Very frequently it happens that the peritoneal mesoblastic epithelium, which lines the perianal ciliated belt, is detached from the ectoblastic wall. This is a purely artificial appearance caused by the killing reagent. It seems probable that MASTERMAN has erroneously considered the space thus formed by splitting to be a vascular space (the "perianal sinus"). The same author states, though with much reserve, that he has discovered a third pair of nephridia in this trunk cavity, which is considered to be a modified part of the body-cavity, and also to be rudiments of the adult nephridia. I can at present say no more than that these are certainly absent in every type of the Actinotrocha studied by myself.

# 2. Organs of Ectoblastic Origin.

The epidermis of Actinotrocha is represented by a single layer of cubical or cylindrical cells, those of the collar wall and of the upper and the lower walls of the hood being provided with well developed cilia. Besides, there are three specially ciliated regions: the preoral belt, the tentacles, and the perianal belt. The last is the larval locomotory organ: on it the cilia are very long, thick, and somewhat bristle-like when in active motion. At places, where cilia are strongly developed, (e.g., the nerve ganglion, the sensory spot if present, the ciliated belts, etc.) the constituent cells are cylindrical, the nucleus generally lying near the basal end. The body wall of the trunk region is very thin and is formed of greatly attenuated cells (especially slender in the advanced larvæ).

Numerous unicellular glands are found in the Actinotrocha not only all over the two surfaces of the preoral lobe, but also in the osophageal wall as well as in the inner ectoblastic wall of the ventral ponch. They are also, though less abundantly, distributed over both the collar wall and the tentacular wall. The glandular cells are all pear-shaped, the nucleus being found always appressed to the base of the cell (figs. 49 and 64 d, m.gl.). In their staining reactions, the secretory contents of the glands agree with those of mucin. It has been often noticed that living larvae remain adhereing to the objects they have touched with the hood, and that metamorphosed larvae behave similarly with the tip of the evaginated pouch.

There exists still another, paired, multicellular gland which is observed only in the larvæ of type C (figs. 15, gld. and fig. 15 c). It is situated on both sides of the median line on the upper surface, and somewhat near, the neck of the preoral lobe. It has the shape of a round flask with a short neck (fig. 15 c). The appearance of the section through the body of this gland reminds us of the chorda dorsalis in Vertebrate embryos: it presents to view a mesh-work of protoplasm, a small number of nuclei being found here and there closely pressed against the reticular beams or the nodes of these (figs. 56 a-c).\* Each of the meshes corresponds to one gland cell. In fig. 56 b, which shows an oblique median section of the body of the gland, a comparatively wide round space exists in the centre, surrounded by the gland cells which are arranged more or less radially. This space, when traced upwards, passes into a short and very narrow tubular canal, finally to lead to the exterior by a small aperture (Fig. 56 c). Since

<sup>\*</sup> By an unfortunate oversight, Fig. 56 b has had its number omitted in the plate.

the neck portion of the gland is very short, it is difficult to prepare a good longitudinal section of it, in which the canal may be seen opening to the exterior. Fig. 56 c represents the terminal part of the emptying canal, which, as can be ascertained by regulating the focus, leads to the external pore. Mr. IIZUKA tells me that similar glands of ectoblastic origin are constantly found on the superior ramus of a parapodium in certain *Polychetta*.

Ventral Pouch. As the ventral pouch is one of the most characteristic structures of Actinotrocha, its form and fate have been fully studied by many previous observers. In the 8-armed larva of type 1, an ectodermal thickening below the tentacular row represents the origin of the pouch. At the 10-armed stage the thickening becomes more conspicuous, but no invagination has as yet taken place. For the first time in the 12-armed stage, the wall at the thickening begins to sink inwards and backwards (fig. 45,  $p_{\theta}$ .). The invagination is lined with a mesoblastic layer, and, as before noted, is for its whole length suspended by the ventral mesentery, joining it to the somatic and the splanchnic walls. As the growth of the larva advances, the pouch becomes more clongated and bends on itself around the alimentary canal )figs. 18 and 57 a, po.). In fully developed larvae of whatever type, the inner or ectoblastic wall is thrown into small wavy folds (beginning at the distal portion near the pouch pore), while the mesoblastic layer becomes muscular, so that at the end of larval life, it forms a thick muscular sheath whose constituent cells stand vertically to the inner wall. As to the form and position of the pouch pore, I can offer no details in addition to what has been observed by Metschnikoff and many other authorities.

Nervous System. The nervous system of Actinotrocha, like that of Phoronis, is of a very low development, being represented

550 1. IKEDA:

merely by a local differentiation of the ectoblastic cells into nervous elements. The epidermis over both the gauglion (fig. 14. gl.) and the sensory spot (so.) is strongly ciliated, so that the organs are easily recognizable in the living larva. The earliest stage in which I found the gauglion was a 4-armed larva of type 1 (fig. 40 gl.). In it, the gauglion consisted of only a few gauglion cells and nerve fibres.

Although the ganglion and some nerves directly proceeding from it can be detected with tolerable distinctness in the living specimen on account of their peculiar refractivity, the peripheral nerves are as a general rule so very fine and delicate, that they can not be satisfactorily made out by means of any ordinary process. With fair success I have had recourse to vital staining with methyl-blue. Larvæ of type B have been principally employed for this purpose. They are left for about 15-20 minutes in a weak solution of methyl-blue in sea water and immediately afterwards treated with ammonium molybdate. Sometimes, I have made supplementary observations on larvæ lying alive in the methyl-blue solution under the cover glass, but this can be continued for only a short time, since a general overstaining of other tissues soon takes place.

Fig. 60  $a^{\circ}$  shows the dorsal view of the anterior half of a larva of type B, which was treated in the above way. The nerves are shown in blue. The results obtained as to their distribution differ in many important points from those obtained by Masterman. Whether this difference is due to the technique or is actually existant in the species studied, is difficult to ascertain.

<sup>\*</sup> As the larva shown in this figure was compressed by the cover-glass, the rim of the hood which appears like its free margin is in reality the line along which the hood was bent and reflected over by pressure. The line drawn close to the peripheral dots in blue represents the true edge of the hood.

MASTERMAN says nothing of the method employed in his investigation, and unfortunately there exists no other study than his with which to compare my results.

As may be gathered from the above-mentioned figure, I can discover no collar nerve ring, nor dorsal or ventral commissure. Besides, in spite of repeated efforts, I have always failed to make out the presence of the so-called perianal nerve ring. The collar ring and the dorsal commisure, if they can be so named, are represented by a small number of parallel fibres, which spring directly out from the posterior corner of the nerve ganglion. case examined, they could be traced no further than a short distance from the ganglion. Sometimes, I have been able to discern in sections the main nerves (commonly 3 in number), which run close together and parallel to one another along the mid-dorsal line of the trunk, but they were confined to only a few sections posterior to the first pair of tentacles. On the other hand, a very complex and beautiful system of nerve fibres could be seen on the preoral lobe. The fibres are here exceedingly numerous and fine, radiating from the ganglion on all sides towards the free margin of the preoral lobe. In the median line and anteriorly to the ganglion (gl.), the fibres appear as three longitudinal parallel strands on which the unpaired sensory spot (so.) is situated not far from the ganglion. After passing through the sensory spot the strands fray out into fine fibres which continue their course towards the free margin of the preoral lobe. The fibres emanating from the ganglion do not all show a regular radial arrangement, but there are some that arising from the lateral edge of the ganglion, soon take an anteriorly directed course. Sometimes there were not wanting, especially near the ganglion, indications of anastomosis between the fibres. However, it seemed 552 1. IKEDA:

to me more probable that these appearances were eaused simply by the juxtaposition of intersecting fibres.

The nerve endings in the preoral ciliated belt deserve special notice. In fig. 60 a, there is shown a row of small dots along the margin of the band. A portion of the latter more highly magnified is shown in fig. 60 b. Here each fibre ends in a small knob which is devoid of any lateral process. At first sight under low magnification, the row of knobs appears like a deeply stained ring. Suspecting that there might exist lateral processes connecting knobs, I have repeatedly made observations and experiments, but without having ever been able to demonstrate such a connection between them.

I can not but think it very strange that post-ganglional nerve fibres, if such really exist in the forms of the collar ring and of the dorsal and the ventral commissures, should not be revealed by the method adopted. The negative result may be considered due to incomplete development of nervous elements in the collar and in the trunk region; but other anatomical relations prove to a certainty that the larvae investigated were fully grown. As I am not quite sure that my method was not in some respect imperfect, I leave the matter undecided for the present.

According to Masterman, there is an ectodermal depression directed inwards and backwards, just in front of, and under, the ganglion. He calls it the "neuropore," comparing it to the neuropore of Amphioxus and even to the medullary canal of Vertebrates. I must say I was much disappointed in failing to detect in the Actinotrochus studied by me this structure of so much theoretical interest. As a matter of fact, it happened very frequently, while observing living larve, that the ganglion was retracted deeply inwards by an active contraction of the two

retractor muscles in the collar cavity (figs. 13, 14, 15, rel.) producing at the same time a deep depression just in front of the ganglion. It is also of almost constant occurrence that the ganglion is withdrawn inwards on the application of reagents, so as to produce a shaflow pit or groove in front of, or below, the ganglion (figs. 63 a, gl.). A quite similar fact is always observed in Lovéx's larva. From these circumstances I am much inclined to regard the "neuropore" of Masterman not as a really existing structure, but as an artefact.

As to the tentacles, I have at present nothing to add to what is already known about them.

# 3. Organs of Entoblastic Origin.

In the fully grown larvæ the alimentary canal is a long and straight tube; it begins with the mouth which is overhung by the preoral lobe, and ends at the anns in the centre of the anal cone surrounded by the perianal belt (figs. 12-16). Of the whole alimentary tract three parts may be distinguished: the Oesophagus, the Stomach, and the Intestine.

Ocsophagus. In the embryological part of this article I have said that the esophagus of Actinotrocha is of ectoblastic origin, so that the original gastrula mouth is to be sought at the juncture of the esophagus with the stomach. The esophagus (Figs. 45, 48, and 49, \(\sigma s.\)) is a comparatively short and narrow canal with a wall composed of densely ciliated cylindrical cells, among which are scattered numerous unicellular glands (m.gl.). Thus the wall does not differ in structure from that of the hood or of the collar.

Masterman has described an unpaired ectodermal invagina-

tion situated in front of the mouth and just under the ganglion. It is called the "subneural gland." Here again I am not in a position to confirm his view. In spite of repeated examinations on living specimens, I have been unable to discover any structure which has the slightest resemblance to the subneural gland. To judge from my own observation, the "subneural gland" as well as both the "oral-" and the "pharyngeal grooves" of this author are products of his fixing method. In preserved specimens, it is frequently noticed that the lower wall of the hood is bulged out and downwards in front of the mouth (fig. 16, prom.), and, as a result of this, there is brought about on the wall behind the prominence a depression, which appears on sections as a tolerably deep pit (fig. 63 a).

Stomuch. The stomach forms the largest and widest portion of the alimentary canal. It is especially long in the larvæ of type D, in which it extends below nearly to the plane of the perianal belt. The greater part of the stomach wall is composed of evlindrical cells with short cilia whose spherical nucleus is usually situated in the centre of the cell (figs. 45, 48, and 50 a, stom.). But the anterior portion of the wall along the mid-dorsal line and the posterior portion near the intestine greatly differ in their constituent cells from the remaining parts. They consist exclusively of tall ciliated cells which contain elongated nuclei, and are, in a word, of the esophageal type (fig. 45). In the full grown larva, the ventro-lateral portions of the stomach wall form two digestive areas placed in the neighbourhood of the septum. Here the cell boundaries are indistinct and the nuclei are imbedded in a common mass of protoplasm, in which remains of various unicellular organisms are enclosed.

From the anterior end of the stomach a pretty wide and

unpaired diverticulum protrudes itself forwards (fig. 14, dir.). The position of the organ is wholly rentral to the esophagus (as.), and the form is like that of a sac compressed in the dorso-ventral direction (figs. 45, 49, 50 a, and 63 u, dir.). The internal cavity is continuous with the stomach cavity. The roof of the diverticulum in the fresh state generally shows a reddish brown tint. coloration is due to the superposition of the fundamental brownish colour on the hæmoglobin of the blood corpuscles which, in advanced larvæ, overlie the organ in either one, or two masses. The cells which compose the diverticular wall are tall and slightly curved, and are ciliated on the free ends (fig. 50 a, div.) In fully grown larvæ of every type, each cell constantly contains a single small round vacuole in its distal end (fig. 61 b). The vacuoles can not be stained by most of the staining reagents. I have seen them in the diverticular wall of a highly advanced larva belonging to type A, which had already evaginated the ventral pouch; even in this case, they were found only one in each cell (fig. 61 b). The whole of the diverticulum is lined externally with the thin peritoneal layer (see the above figure).

Many previous observers have noticed this organ and have called it by various names:—

J. Muller ('46)—" Blinddarme" (paired),

GEGENBAUR ('54)—"Haufen der Leberzellen."

WAGENER ('47)—" Leberblinddarme,"

CLAPARÈDE ('63)—" A dark mass with globules " (after Masterman).

METSCHNIKOFF ('71)—"brown specks,"

Wilson (A.G.) ('81)—" glandular lobes of the stomach,"

Masterman ('97)-" Notochord" (paired),

Roule ('98)—" Notochord " (unpaired).

556 L. IKEDA;

Thus it will be seen that while some authors have apparently confounded the organ with the overlying corpuscle masses, others have considered it to be a glandular appendage of the stomach, and still others have regarded it as a skeletal structure. According to Masterman, who maintains the last mentioned opinion, the stomach wall is produced, in the antero-lateral region, "into two remarkable diverticula which in the fully developed larva lie as a pair of clongated organs, Notochords, laterally to the œsophagus" ('97, l.c., p. 302). The organs are said to soon undergo a remarkable metamorphosis, i.e., vacuolization. The vacuoles are produced successively one after another at the distal ends of the cells and are arranged alternately in several layers. On account of these facts Masterman rejects the view that the organ is of a glandular nature, and holds that it is to be compared in function and structure with the notochord of the Chordata. In 1898 Roule published his third paper on Actinotrocka, in which he denied that the organ is double in number and lateral in position to the esophagus, but admitted the vacuolization in the larva of Phoronis sabatieri (=P. psammophila Cori).

I can not at present decide whether the variations in the number of the diverticulum and in the degree of vacuolization are of specific value or not. For the present I must be content with simply noting that the stomach diverticulum in the larvæ studied by me is constantly unpaired and undergoes no farther vacuolization process than the production of one vacuole in each cell.

Intestine. The intestine which leads to the anns is a slender canal whose wall is composed of a layer of somewhat cylindrical, ciliated cells with round nuclei (figs. 45 and 48, int.).

## 1. Organs of Mesoblastic Origin.

As the mesoblastic organs have been but little studied in their development, so their structure and fate after metamorphosis are very imperfectly known. Although I have endeavoured to make my study of the organs as exhaustive as possible, some important questions remain yet unsolved. The principal organs to be described in this place are the muscular elements, the vascular system, and the nephridia.

Nephridia. I will treat these under the mesoblastic organs, for, though the nephridial canals are of ectoblastic origin, the organs as a whole bear intimate relations to the mesoblast. Most of the earlier observers overlooked the presence of nephridia in the larva. The first discoverer was Wagener ('48), whose description is, however, very meagre and gives us no exact idea of the organ. Caldwell in his preliminary note ('82-'83) has given a detailed description of the nephridia. According to his view, the nephridial canal at no time during larval life, opens into the body-cavity.

Masterman ('97) has described the exerctory system of the larva in detail and has suggested an hypothesis which seems to me to be an extraordinary one. Each of the three "segments" of the larval body, he concludes is provided with a paired organ which performs the excretory function. The three pairs of organs are called respectively the "proboseis pores," the "collar nephridia," and the "trunk nephridia," Of these, however, the presence of the first and the third is, as I have before pointed out, very doubtful. The second pair, or the collar nephridia, are the organs which I consider to be the nephridia. Masterman's views on the

structure of the nephridial canals are in the main similar to those of Caldwell, except in one important point, viz, that the canals are said to open by means of funnels into the collar cavity.

When a larva of any type is examined in the living state, the proximal ends of the organs are seen, as described by Wagener, as two bonquet-shaped masses which are formed by a crowding together of the excretory cells (fig. 13, neph.). They are placed symmetrically one on each side of the stomach and in front of the postoral septum. Each of them consists of two parts, the nephridial canal and the excretory cells. The former is composed of a layer of cubical cells, and contains a narrow lumen which ends blindly at the internal end and distally leads to the nephridial pore lying on either side of the pouch pore. The greater part of the nephridial canal, together with the excretory cells, rests on the upper surface of the postoral septum. Fig. 50 bshows a cross section of the larva, passing through the left nephridial canal near its internal blind end, where the excretory cells In the above figure, a small cell mass (nep.c.) on the right of the figure, shows the cut end of the nephridial canal which is attached to the septum (mes.). In the figure, the left canal (nep.c) containing a small lumen, is found applied to the somatic walls. If traced a little downwards, these two canals become attached to and imbedded between the two layers of the somatic walls, and are no more to be seen in the collar cavity. Such a state is represented in fig. 50 e, in which the two canals (nep.e.) are wholly imbedded in the somatic walls on both sides of the stomach (stm.). This condition is more distinctly shown in figs. 47 (a-c), which are taken from serial longitudinal sections of a larva of type A with 12 tentacles. These figures show only one portion of the skin, where the nephridial canal and the somatic attachment of the postoral septum (mcs.) are situated. In fig. 47 n, one portion of the peritoneal layer of the stomach wall is also represented. Now we see in the first two figures of the above series, that the nephridial canal (nep c.) which is here imbedded in the somatic layers, lies distinctly below the septum (mcs.). So, in the third figure the nephridial pore (nep.o) is seen as a small pit in the trunk wall, which is situated considerably below the septum. The infraseptal position of the nephridial pores has also been acknowledged by Caldwell. Though Masterman has made no direct statement on this point, it may safely be inferred from his figures, that he must have regarded the pores as lying in front of the septum.

Fig. 51 a represents a longitudinal section through the middle of the supraseptal portion of the nephridial canal. Here the canal appears as a comparatively long tube with a narrow lumen; it is invested throughout with a thin mesoblastic epithelium. At its upper extremity where the lumen disappears, a certain number of spindle-shaped excretory cells is found aggregated together. In fig. 51 b, which is taken from the same series as fig. 51 a, the canal has wholly disappeared from the section, leaving only a bunch of the excretory cells (exc.c.) adhering to the septum (mcs.). All of these spindle-shaped cells have their nuclei in the swollen ends. I have never found either among, or in, the neighbourhood of the cell bunch any perforated excretory cells bearing many processes,—cells which are said to have been present in the Actinotrocha studied by Caldwell.

Masterman considers that each bonquet of the exerctory cells is composed of a cellular mass traversed by a system of minute funnels; and that these funnels communicate with the main canal of the nephridium as well as with the collar cavity. But I may

say with certainty that, at least in the larvæ studied by me, there existed no such funnel-system nor any such free communication between the collar cavity and the nephridial canals. The same negative result was also reached by me in my examination of the just metamorphosed larva of type A. Thus in fig. 64 f which is drawn from a section through the tip of the nephridium, the excretory cells (exc.c.) still remain compactly grouped on the blind tip of the canal (nep.c.), but are not traversed by any sort of canal-systems.

Muscular System. The muscular system of Actinotrocha remains in a low state of development, which may account for the fact that most previous observers have paid no particular attention to it. It was therefore of much interest to discover two pairs of tolerably well developed muscles, which had hitherto remained apparently unknown. They show a strong resemblance to the retractor muscles which have been known in many forms of Trochophora larvæ.

Though the longitudinal and circular muscles of the body wall may be observed with tolerable distinctness in the living larva, they are usually very poorly preserved after hardening. They are all subdermal in position and very delicate in structure, so that as a rule they can not be satisfactorily distinguished from the underlying peritoneal layer. However, in certain preparations of the entire larva the circular muscles of the upper and lower walls of the preoral lobe and of the trunk body wall could be detected as fine deeply stained fibres. In the same way the longitudinal muscles of the collar wall, especially in the larva of type D, were fairly traceable. The larva of that type also exhibited a peculiar arrangement of the circular muscles of the trunk, in that these formed four, equidistant, longitudinal series

around the periphery of the trunk (see fig. 12). In the larva of type C I have always found a comparatively thick layer of In fig. 57 b, which shows a portion of the circular muscles. trunk wall containing the nephridial canal (nep.c.), the muscles are represented as a thin fibrous layer (cir.m.) intercepted between the ectoderm and the peritoneal epithelium. The floor of the mouth just opposite the stomach diverticulum, is always associated with a particularly well developed muscular sheet. The mesenchymatous unicellular fibres which traverse the preoral cavity are to be regarded as a kind of primitive muscles. The most highly developed parts of the muscular system of the somatic, and of the splanchnic, walls are to be found in the muscular sheaths of the ventral pouch and of the dorsal wall of the stomach in the advanced larvae of all types. Each of them is formed of a thick layer of enormously elongated muscular cells which stand vertically to the ectoblastic, or the entoblastic, wall as the case may be (figs. 58 c and 63 e, m.sh.). The sheath of the stomach wall is thickest along the mid-dorsal line of the stomach: it is shown in fig. 58 c and fig. 63 c, the former figure being taken from a cross section and the latter from a longitudinal section through the dorso-anterior region of the stomach. The muscular sheath (or the external wall) of the ventral pouch is essentially similar to that of the stomach.

The Retractor Muscles can be constantly detected in every type of the larva as two slender threads on both sides of the cosophagus (figs. 12, 13, 14, 15, ret.). They spring from the hind lateral corners of the ganglion (gl.) and run divergently downwards until they insert themselves in the collar walls between the first, and the second, tentacles. In order to obtain a clear idea of the position of these muscles, it is necessary to study them in sections:

the larvae of types B and D are best suited for this purpose, as the muscles in these are remarkably large and long. Figs. 63 a-c are taken from serial sagittal sections through the right side of the nerve ganglion. In figure a, a median section, we see behind the ganglion nothing but the preoral septum (mes'.) In the next figure, b, the septum is found to have shifted to a more anterior position and its dorsal termination is accompanied by a strong muscle-band (ret.). This band corresponds to that portion of the retractor muscle which is nearest to its auterior insertion on the postero-lateral side of the ganglion. The two muscles are shown in fig. 59 d (rel.), a cross section through the posterior recess (p.r.) of the preoral cavity, where they spring directly from the septum (mes). In fig. 63 c, which shows a more lateral region than fig. 63 b, the muscle is found to have retreated far backwards, touching with its posterior portion the resophageal walls (es.). The posterior insertion of the muscles on the somatic walls are best studied in serial cross sections of the larva. Figs. 58 u and b show two cross sections passing through the mouth (a) and through the middle of the resophogus (b). In both figures the muscles (ret.) are found on both sides of the esophagus (as.). A little further down they soon detach themselves from the osophagus and begin to traverse freely the body-cavity (larval collar cavity), and after that they again apply themselves to the skin on each side between the first and second tentucles (t' and t'' in fig. 58 b).

There is also present another pair of muscles, which can be discovered only in the larvæ of type C. They are so very long as to equal the entire length of the trunk (fig. 15 b, ret'.). They arise on each side from the somatic walls just above the nephridial pore and run straight downwards traversing the trunk cavity, and ending at the terminal portion of the intestine. Fig. 57 a

represents a transverse section through the middle portion of the trunk where the stomach (stm.) joins the intestine (int.). There the muscles appear as two small striated masses (ret.) lying on both sides of the intestine. As represented in fig. 57 b, which is taken from the portion of the body wall containing the nephridial canal (nep.c.), each of the muscles is in its origin traceable to the circular muscle layer (cir.m.) which is subdermally interposed between the ectoblast and the peritoneal epithelium. When these two muscles, in their downward course, reach the level of the terminal portion of the intestine, they fuse together into one on the dorsal side of the intestine. I think that the above mentioned muscles are a kind of retractors serving to contract the trunk of the larval body. I can not help thinking that the "Afterbänderung" of Wagener ('47) is probably simply the posterior portion of the nuscles in question in the proximity of the anus.

Vascular system. It is a well known fact that the closed vascular system of *Phoronis* offers one of the greatest obstacles to the idea entertained by some naturalists that the animal is of the Polyzoan type. Many writers are, therefore, much inclined to attribute the simple body organization of *Phoronis* to secondary adaptation, and to erect the animal into a distinct order very closely related to the Chordata. Putting aside for the present all theoretical speculations, it is of great importance in ascertaining the phylogenetic relation of the animal to note that one portion of the larval body-cavities is transformed into a blood vessel, and that the simple and rudimentary vascular system of *Actinotrocha* undergoes a wonderful change and suddenly attains the high organisation seen in the adult during metamorphosis.

Krohn ('50) proved that the "Leberzellen" of Wagener and Gegenbaur were really blood corpuseles. However, he did

not discover any blood vessel in Actinotrocha: he thought that the blood vessels of the metamorphosed worm arose in the corpuscle masses of the larva.

CLAPARÈDE ('63) mentioned a ring-like vascular canal under the tentacular row of the larva, but did not explain its nature.

Schneider ('62) discovered two vessels in Actinotrocha, which ran parallel along the mid-dorsal line of the stomach.

Metschnikoff ('71) described and figured in a larva of 10 tentacles the "feinen Häutehen" situated just above the invagination pouch, which was said to be the "Gefässanlage," Besides, it is stated that he saw a ventral "sinusartigen Schlauch" which covered the greater part of the stomach and communicated anteriorly with the collar cavity. According to his view, this Schlauch should give rise to the ring vessel of the adult. But what are really meant by the "Schlauch" and the "Häutehen" is not clear from his text and figures.

Wilson ('81) confirmed the main points of Metschnikoff's observations, but disproved the presence of a blood vessel along the intestine, and also the free communication between the pseudo-hæmal space and the perisviceral cavity. According to this author, there are two sorts of corpuscles: the one kind floats in masses in the perivisceral cavity, and the other (the pseudohæmal corpuscles) arise within the cavity of a sinus which is formed in the stomach walls and form the circular ring vessel of the adult.

Caldwell (82-83) gives us a concise description of the vascular system in Actinotrocha and in its adult form. He says that the corpuscle masses "arise from the mesoblast cells in front of the septum," and that "The vessels arise as slits in the splanchnopleure. The adult condition is reached partly by constrictions, partly by out-growth from these. Thus we have at the

close of the larval life the blood system in the following condition:

- 1. Blood corpuscles aggregated in two or more masses, lying in the body-carity of the preoral lobe, i. e., in front of the septum.
- 2. A blood ressel formed on the dorsal wall of the stomach, a marked structure of the larra.
- 3. The splanchnopleure, which in the region of the stomach forms a loose suc surrounding the gut.
  - 4. Creal prolongations of this sac.
- 5. Coveal prolongations into the rudiments of the adult tentacles" ('82-'83, l.e., p. 377). Besides, the author insists on the free communication between the splanchnopleuric sac and the body-cavity in front of the septum. Thus it may be understood that Caldwell detected only one vessel (dorsal) in Actinotrocha and thought the ring vessel of the adult was produced from the splanchnopleuric sac around the stomach.

Masterman's views ('97) of the vascular system differ greatly from those of all the others above quoted. The subneural sinus is said to communicate posteriorly by a chink with the dorsal vessel on the osophagus. The dorsal vessel runs down till it communicates with the ventral vessel at the juncture of the stomach and the intestine, by means of a small ring sinus. Anteriorly also the dorsal vessel gives off two branches which, after passing along the inner side of the two notochords, again meet together in the mid-ventral line, forming a post oral ring sinus. From that meeting point originates the ventral vessel which runs down along the whole length of the gut and opens into a large sinus-ring situated just within the perianal belt. Further, the author denied the free communication of the blood vessels with the body-cavity, which had been maintained by Metschnikoff and Caldwell.

I have already stated my belief that the mother cells of blood

566 1. IKEDA:

corpuseles appear as the gigantic mesoblast cells in the body-cavity of the larva with one or two pairs of tentacles (fig. 44, corp.). These cells may be easily distinguished from other wandering mesoblast cells in that they have an enormous size and are loaded with an abundant quantity of large volk grains. Now in 8-10-armed larvæ of type 1, not only such peculiar cells but also the so-called corpusele masses can not be found in any part of the bodycavities. Instead of them the collar cavity and often also the tentacular cavities contain a few large and isolated mesoblast cells which closely resemble in size and structure the blood corpuscles of a highly advanced larva. These cells no longer contain large yolk grains, but enclose numerous fine, refringent granules. Fig. 46 represents two such corpuscles (corp.) floating in the tentacular cavity (t.) of a larva of 10 tentacles. It can with propriety, I believe, be admitted, that these corpuscles have arisen by repeated division from the gigantic mesoblast cells, whose yolk contents have been gradually used up during the process. If this be not the case, then how is the presence of those isolated corpuscles in the young larvæ to be explained? If, as is imagined by Caldwell, they are produced by cell-multiplication taking place in certain parts of the splanchnopleuric walls and form the corpuscle masses from the first, why should such freely floating and isolated corpuscles be actually present in the young larvae in which the masses are not yet discernible? So far as I know, in the larvæ of type 1, the corpuscle masses do not exist until the animal has so far developed as to possess at least 14 tentacles (fig. 13, corp.). At the stages of 14 and 16 tentacles, these masses are present commonly in two pairs, the one covering the stomach diverticulum and the other just in front of the septum and on both sides of the stomach (fig. 13, corp.). They appear as pinkish

spheres in fresh specimens. They do not constantly adhere to the stomach walls as Wilson and some others have remarked, but are very frequently found floating freely in the collar cavity, showing that there exists no direct connection with the splanchnic walls. This fact may be clearly seen in fig. 53 (corp.), where the mass is located at an appreciable distance from the stomach walls (stm.). The same state was also observed in a 14-armed larva of the same type. Besides, I have noticed at these stages of growth two sorts of corpuscles in the masses: the one sort is large and somewhat coarsely granular; the other is much smaller and finely granular. On sections it was found that the former sort is imbedded here and there in groups of the latter (see fig. 53, corp.). I can not exactly see the significance of this fact unless it be that it shows the developmental process of the blood corpuscles, in which the larger ones give rise to the smaller by division (the karyokinetic figures in the former can be made out with tolerable distinctness by staining with cosin methylblue). The larger cells are essentially identical with the corpuscles of both the vounger (12-armed) and the older larvæ (so far as advanced as to be ready for metamorphosis) of the same type. Thus it seems to me probable, though I state this with a certain degree of reserve, that these smaller corpuscles develop into the normal blood corpuscles of the highly advanced larva, for in the latter we no longer find the smaller forms in the corpuscle masses. Fig. 61 a represents four corpuscle cells composing a corpuscle mass of a larva of type A which has already evaginated the ventral pouch. Of course at such a stage approaching the end of larval life, the number of the corpuseles is actually greatly increased as compared with that in vounger stages.

From the above observations it may be concluded that the

blood corpuscles of Actinotrocha do not arise at the expense of the splanchnic walls, but are produced by a continual division of certain previously differentiated mesoblast cells.

I will next describe the blood vessels which can be seen during larval life, with reference to the formation of the adult collar cavity. I have shown in the foregoing pages that Master-Man's subneural sinus is probably nothing but a posterior recess of the preoral body-cavity, and that neither the dorsal vessel nor the dorsal mesentery is present on the cosophagus in any species of Actinotrocha I have been able to obtain. I have endeavoured to ascertain the presence of the dorsal and the ventral vessels as well as of the ring-sinuses around the gut, and I am convinced that at no time during larval life any vessels other than the dorsal on the stomach and the cocal capillaries are present in the larvae.

In the A type-larva of 14 arms, the dorsal vessel, as figs. 50 b and 50 c will show, is not yet formed and the stomach wall is uniformly lined with a thin mesoblastic layer. This layer thickens later and its constituent cells become muscular, beginning first at the base of the postoral septum and along the mid-dorsal line. When the larva grows to the stage of 16 tentacles, the dorsal vessel is inceptionally formed. It arises as a solid cord of cells interposed between the muscular, and the entoblastic, walls of the stomach. As shown in fig. 52, the vessel in section is represented by a loose mass of mesoblast cells distinctly delimited on all sides from the surrounding parts; but as yet no lumen is visible in it. I have not seen the definite lumen establish itself in this rudiment of the dorsal vessel at any time during the whole larval life of this type, while, on the other hand, in the advanced larvae of the other three types, it could be readily recognized as such.

Fig. 58 c represents a portion of a transverse section through the anterior region of the trunk of a larva belonging to type D. The vessel in question here appears as a small canal (d,r) running in the stomach walls (stm, and m.sh.). As will be seen in the figure. the canal is distinctly lined with an epithelial cell-layer. dorsal vessel terminates anteriorly just behind the postoral septum, so that the whole course of the vessel is confined to the trunk region. During larval life, the dorsal vessel does not extend so far posteriorly as to become confluent with the corcal contractile capillaries which are formed at the point of juncture of the stomach and the intestine. Thus we see in Fig. 55b, which is taken from a transverse section through the lower portion of the stomach, that the gut is covered with a thin mesoblastic wall without a trace of the dorsal vessel, but the capillaries (r.c.) are here already developed at this period. In the above montioned figure they are found as cell masses protruding into the trunk cavity from the right side of the splanchuic attachment of the ventral mesentery to the gut; one capillary is seen in cross sec-The capillaries shown in that figure are certainly in an early state of development, and, when fully developed, they appear like a tuft consisting of tolerably long, blindly ending tubes. Sometimes I have observed that the capillaries are formed not only on one side of the ventral mesentery, but on both sides of it; and that they are not constantly formed on the gut walls, but sometimes on the ventral mesentery. Thus we see in fig. 57 a on the ventral mesentery (r.mes.) a rosette-like figure (r.c.), the rudiments of the capillaries seen under a low power. It is represented highly magnified in fig. 57 c (taken from another neighbouring section of the same series). Here are seen signs of cell-multiplication on either side of the ventral mesentery (r.mes.), in places

not in direct contact with, but separated by a considerable distance from, the gut.

From the facts above stated, we are justified in concluding that the ceed capillaries are not produced as out-growths of the dorsal vessel, but are formed independently by cell-multiplication taking place in certain parts (near the gut) of the ventral mesentery, and, therefore, that the dorsal vessel and the capillaries have different origins.

Next I will consider the origin of the ring vessel of the adult animal. Although some early authors have frequently referred to the so-called ring vessel of Actinotrocha, yet its origin, form, and position have never been satisfactorily elucidated. Nobody has investigated it by means of sections, and the statements which have been made about it do not rest, it seems to me, on actual anatomical studies of Actinotrocha, but rather are mere inferences from facts known respecting the metamorphosed larva. Consequently there have been put forth several irreconcible views in regard to the ring vessel. The structure described under that name by Claparede, Schneider, Metschnikoff, and Caldwell does not seem to be even one and the same thing.

In order to make clear the relations of this system of organs, I must first of all describe somewhat minutely the adult collar cavity. In the fully developed larva of every type, the rudiment of that cavity is represented by a circular space on the inner side of the tentacular row just above the septum. The space is in form not a complete ring, but is interrupted at the median

<sup>&</sup>quot;In passing I should say that MASTERMAN's ring sinuses have nothing to do with the vessel under consideration, because of them the first is stated to be priosophageal, the second peri-intestinal, and the third perianal in position; and thus they must be something entirely different from the ring vessel of other authors, which is a sinus in the splanchnic walls around the stomach.

dorsal point. The relative position of it with regard to the septum and the tentacles can be most conveniently studied on sagittal sections of the larval body. In fig. 63 a the adult collar cavity (s.c.c.) is indicated by a vertical club-shaped space just inside the the body walls and above the postoral septum (mes.). Fig. 63 dshows a portion of the ventral part of a nearly median sagittal section similar to Fig. 63 a. Here the cavity (s.c.c.) is seen also as a vertical and comparatively wide space situated inside the body walls and below the tentacles (p.t.). It can also be made out that the wall of the cavity is formed of a single layer of mesoblastic cells and its ventral wall is in contact with the somatic walls of the adult tentacle (s.t.), while the posterior wall is superposed on the anterior side of the septum (mcs.). In fig. 58 a and 58 b the two cellular circles (s.c.c.) attached to the inner side of the tentacles (t'.) represent a somewhat obliquely cut transverse section of the adult collar cavity. The two tentacles belong to the first pair, and the farthest dorsal point reached by the cavities is, therefore, at the bases of these tentacles. When the serial sections are traced posteriorly, these cavities gradually extend more and more ventrally along the body walls and at last join with each other in the median ventral line. In fig. 58 c the eavities appear as two narrow spaces (s.c.c.) appressed against the ectoblast (ech). In fig. 55 a, which is taken from a cross section of an A-type larva cut nearly parallel with the tentacular row, the eavity (s.c.c.) appears as a long slit-like space intervening between the postoral septum (mes.) and the body walls. Here the cavity is seen entirely free of the septum, because the section passes through that portion of the collar which lies slightly above the somatic insertion of the septum (compare fig. 63d).

In somewhat younger larvae of all types, the adult collar

cavity is not yet extended dorsally as far as in fig. 58. Thus, in fig. 63 e it is represented by a mesoblastic cell-mass (s.c.c.) which is placed just under the second tentacle (t."), and encloses no lumen in itself as yet. It is evident therefore that the adult collar cavity extends itself during development from the ventral towards the dorsal side of the body, as is also the case with the tentacles.

As the larva reaches the end of the swimming period, the adult collar cavity in the adult tentacles becomes wider and wider, nearly filling up the interior of the latter, while the larval tentacular cavity is henceforth gradually reduced to a narrow space appressed to the upper roof of the tentacle. Fig. 58 d represents a median sagittal section of a larval tentacle of a larva of type D. The portion belonging to the adult tentacle (s.t.) is characterised by a very thick ectoblastic layer forming the ventral wall of that tentacle. The adult cavity appears as a remarkably wide space (s.c.c.) beginning at the somatic insertion of the postoral septum (mes.) and ending at the tip of the adult tentacle (s.t.) The narrow cellular band (p.c.c.) resting upon the adult cavity (s.c.c.)corresponds to the larval collar cavity of the tentacle. The larval cavity is clearly seen in cross sections of the tentacle; in fig. 58 e it is visible as a small space (p.c.c.) inclosed by the adult cavity (s.c.c.) except at the median dorsal point. Tracing the cavity (p.e.c.) in the above figure to the base of the tentacle, we see that it communicates by a tiny opening with the larval collar cavity. That portion of the tentacle, which is thrown off during the metamorphosis (see fig. 58 d) is distinctly different from the persistent portion (the adult tentacle) in that the former has no trace of the adult tentacular cavity (s.c.c). I have ascertained after repeated examinations, that the retrogressing larval tentacular

cavity becomes after metamorphosis the tentacular vessel of the adult, as will soon be further explained.

Concurrently with the growth of the adult collar eavity the larval cavity reciprocally diminishes in extent, and finally after metamorphosis it is reduced to a narrow cavity surrounding the gut in front of the postoral septum: this is the ring ressel of the adult. In fig 65, which is taken from a transverse section through the tentacular region of a larva (type 1) just undergoing metamorphosis, a semicircular space (cir.c.) on the left side of the gut represents the greatly reduced larval collar cavity or the ring vessel, which is found dorsally attached to the somatic walls and is laterally quite independent of the latter, though ventrally it is fused with, and rests on, the postoral septum (mes.). One of the tentacular vessels which proceeds from the ring vessel (cir.c.), is denoted in the plate by t.r. Fig. 64 e also shows a transverse section of the head portion (or a frontal section through the supraseptal portion) of another partly metamorphosed larva represented in fig. 11. Here a comparatively spacious cavity (cir.c.) surrounding the gut and filled with the corpuscles, represents the ring vessel from which the tentacular vessels are given off, though this is not shown in the figure. In the above two figures the adult cavity distinctly appears as a narrow space (s.c.c.) outside of the ring vessel (cir.c.). In these stages of the metamorphosis, the both dorsal ends of the adult collar cavity become continuous with each other so as to form a completely circular space above the postoral septum.

I am still uncertain as to how this rudiment of the ring vessel comes into communication with the dorsal vessel or with other vascular spaces which make their appearance during metamorphosis, for it is almost impossible to obtain larvae of intermediate stages 574 1. IKEDA:

in which this vascular communication is just becoming established. But from observations on metamorphosing larvae I have obtained certain suggestions respecting this process.

# III. Metamorphosis.

As to the external changes of the larval body accompanying metamorphosis. I have scarcely anything to add to the exact and detailed descriptions given by Metschnikoff and Wilson. I will, therefore, confine myself mainly to some anatomical points which have been less studied by previous observers. My observations of the metamorphosis were mostly made with the larvae of types A and B which were most abundant in the neighbourhood of the Station. Sometimes I have observed under the microscope the whole course of the phenomenon, the duration of the so-called critical moment being usually not more than 15–25 minutes.

Among the the material obtained with the surface-net we often find larvæ which carry about the partly evaginated pouch, but these individuals can not be said to be undergoing metamorphosis in the strict sense of the term, for they may continue the free swimming life for several days after capture. Besides, they do not show any remarkable change in the internal organs. In them the corpuscles are still in masses, the nephridia preserve their original form and position, while the alimentary canal is of the ordinary form and length.

When the metamorphosis takes place, the partly evaginated pouch protrudes suddenly outwards to its full extent and the alimentary canal is thrown into convulsive contractions. Meanwhile the latter, especially its esophageal and intestinal port-

ions, is drawn out into a long tube, and at the next moment the junction of the stomach and the intestine is first of all pushed into the now spacious pouch. Thus, the anus and tentacular region are brought into close approximation on the dorsal line of the trunk; the digestive canal folds back on itself into an U-shaped tube, as we find it in the adult. The larval tentacles and the preoral lobe are cast off and digested in the stomach; the perianal ciliated belt atrophies in situ. It is during this moment that the corpuscular masses break away and their elements are scattered in the blood vessels, some of which are then being formed. It is highly interesting to observe the breaking up of the corpuscular masses, and the motion of the corpuscles in consequence of the rhythmical contraction and expansion of the dorsal vessel and of the cocal capillaries. The deep pinkish colour of the corpuscles makes it easy to observe their progress into the vessels. Ordinarily after about twenty minutes the vascular system of the worm is completely formed and the circulation characteristic of Phoronis can be noticed. The skin of the foot which now forms the principal part of the body, becomes more opaque on account of the secretory products from the innumerable unicellular glands.

To understand the details relating to the metamorphosis we must examine the animal in sections. Figs. 61 (a-d) show a discontinuous series of cross sections of the larva represented in fig. 11, which is approximately at the critical moment of metamorphosis. I will briefly sketch with the aid of these figures the general internal change during metamorphosis.

Entoblastic Organs. When the ventral pouch is fully evaginated and the stomach is pushed into the pouch, the esophageal portion elongates downwards enormously, so that the stomach diverticulum descends far below the postoral septum, that is to say, 576 1. IKEDA:

into the infraseptal cavity (fig.  $64 \, r$ , dir.). The vacuoles in the cell of the diverticular wall disappear at this period, and the diverticulum itself is immediately afterwards wholly obliterated, probably as the result of a histological atrophy. The stomach wall does not essentially differ in structure from that of the larva.

Mesoblastic Organs. Among the mesoblastic organs the vascular system undergoes most noteworthy changes. Mastermax has maintained that this forms a completely closed canal system even in the free swimming stage of the larva. So far as I have been able to ascertain, the closing up of the vessels into a continuous system occurs after the critical moment of metamorphosis. Having already described my own observations respecting the origin of the ring vessel of the adult. I will now describe other vessels which arise during metamorphosis.

Fig. 64 a shows a cross section of the foot near the posterior extremity where the alimentary canal is bent upon itself. Here we see the cut ends of three contractile capillaries (r.c.) and three simises (s,s) in the stomach walls. A comparatively wide space (d.r.) is found intercepted between the two limbs of the alimentary canal. This space corresponds to the most posterior portion of the dorsal vessel, which, if traced further posteriorly, shows itself to be continuous with both the capillaries (r.c.) and the sinuses (s.s.). A short distance more anteriorly, the dorsal vessel divides into two parts each of which attaches itself to a limb of the alimentary canal; still more anteriorly the branch on the intestine disappears. In that portion of the stomach which lies close to the asophageal tract, the dorsal vessel becomes enveloped with a thick muscular sheath which we have before seen in Actinotrocha (fig. 64 b, d.v.). The vascular sinuses gradually tend to unite into one common space lying on the ventro-lateral side of the gut (fig. 64 b, s.s.; fig. 64 c,

r.r.). At the place where the degenerating stomach diverticulum still persists, the sinuses completely blend together into one large blood vessel corresponding to the ventral vessel of MASTERMAN (fig. 64 c. v.r.). This vessel acquires a definite form at a more anterior region of the esophagus, becoming lined on its sides with a thin mesoblastic wall (fig. 64 d. v.r.). At about this level the dorsal vessel (d.v.) on the esophagus becomes a small canal, such as we know it to be in the adult animal. According to my observations, the large ventral vessel opens at this stage not by two branches, as is the case in the adult, but by one directly into the ring vessel. To my great regret, however. I have not been able to study microscopically the larvae in which the communication between the ring vessel and the dorsal or ventral vessel was in the process of being established.

Now we see that the dorsal vessel of Actinotrocha corresponds to the afferent, and the ventral vessel to the efferent vessel of the adult. The sinuses around the stomach, which have newly arisen during metamorphosis, develop into the complicated organ of the adult.

Nephridia. At the stage when the metamorphosis takes place, the nephridia do not show any important alteration in form and structure from those of the swimming larva: the excretory cells (exe.e) are found still attached to the blind end of the nephridial canal (fig. 64 f, nep.e.) and the external nephridial pores open behind the septum (fig. 64 e, nep. | o.). In fig. 64 e we notice only that the nephridia as a whole have shifted to a more dorsal position than that occupied in the preceding stages (compare with fig. 50 e). This shifting of position becomes more and more marked as the metamorphosis advances, so that when the process is nearly finished, the nephridia on both sides come close

to the so-called anal ridge on the dorsal side. Thus we see in fig. 66, which shows a transverse section through the nephridial region of a completely metamorphosed larva of type A, that the two nephridial canals (nep.c.) are situated very close to the intestine (int.) which lies in the dorsal median line, and that one of them (the right in the figure) opens to the exterior. By examining serial sections it was found that the excretory cells were entirely absent on the nephridial canals, and that the latter were of an inconsiderable length, ending blindly at the inner extremities. It seems very probable, as was pointed out by Caldwell, that the exerctory cells of the larval nephridia are thrown off into the bodycavity; it is also probable that that portion of the nephridial canal, which lies in the collar cavity of Actinotrocha, is obliterated, since, as we see in fig. 66, the inner end of the canal (the right in the figure) lies wholly outside of the septum (mes.), that is to say, in the trunk walls, as is known to be the case in the adult. Thus we may assume that the formation of the infraseptal nephridial funnels of the adult is due to secondary outgrowths of the infraseptal portion of the atrophied, larval nephridial canals.

Ectoblastic Organs. As Caldwell has correctly observed, the larval tentacles and the greater part of the preoral lobe are torn off and are digested in the stomach. I have very often met with the remains of these organs in the interior of the latter (R, fig. 61a). Although I have said, in agreement with Caldwell, that the greater part of the preoral lobe is cast off, yet I can not agree with the view entertained by several authors, that the epistome of the adult developes from the remnant of the preoral lobe of the larva. Because, according to my studies, the nerve ganglion of the larva, which nearly marks the posterior limit of the lobe, can not at all be discerned in the larva at such a stage as is

represented in fig. 11. It is no doubt thrown off together with the other parts of the preoral lobe. Besides, it is equally true that the epistome can not be found in the neighbourhood of the larval month at such a stage, while, on the other hand, in the worm after metamorphosis the rudimentary organ is seen as a small bud on the dorsal median margin of the month.

As before noted, the ring nerve of the adult is not yet formed in the swimming larva. This nerve and the so-called brain of the adult are of new formation: and the complicated nervons system which had developed only in the preoral lobe, suffers the same degeneration as that larval organ. In fig. 64 e the ring nerve  $(r.n.)^{\oplus}$  is seen in section, just exterior to the septum (mes.). It consists of a thick layer of very fine nerve fibres.

# IV. Supplementary Notes.

J. MÜLLER ('46), the first discoverer of Actinotrocha, described the animal as an adult worm under the name Actinotrocha branchiata. The ventral pouch was considered by him to be the sexual organ. Doubts were afterwards thrown on his views by Krohn ('54); and they were finally refuted by Schneider ('62), who maintained that Actinotrocha is the larval form of a certain Gephyrea. This idea was confirmed by Kowalewsky ('67) who ascertained that Actinotrocha is the free swimming larva of Phoronis. Since this renowned discovery of Kowalewsky numerous papers on the anatomy and development of Phoronis have been published by many celebrated naturalists. But the singular fact is that the life history of the animal has not been subjected

<sup>#</sup> Unhappily the letter r is misprinted in the plate as r.

to a detailed study. The metamorphosis of Actinotrocha is, of course, one of the most curious phenomena in the animal ontogeny. But the question which interested me almost to the same extent was: how do the free swimming larvae come to establish colonies at such fixed and limited spots as are found in the Aburatsubo inlet? Accordingly I made several visits to the Misaki Station solely for the purpose of obtaining clues to the elucidation of this point. The results obtained are, I think, worth mentioning.

As I have before stated, the breeding season of *Phoronis* ijimai ranges through about one half of the year, say, from November to June or July, during which months the swimming larvæ, though few in number, are constantly found in Aburatsubo. They, are, however, most abundant from the middle of July to the middle of August. Among these larvæ some are very young, having apparently just swum out of their birth-place; but the majority of them are fully grown.

On July 16th., 1898, I visited the place where *Phoronis* ijimai flourishes, to see if it was still in possession of embryonal masses. But these could no longer be found in the tentacular coils of the mother animals which were, however, in the normal state.

On the 22nd, of the same month, I went again to the same place, and every thing was in the same condition as before.

On August 6th. I visited the place for the third time and to my astonishment I discovered that the animals had all died off. Several deformed colonies were brought back to the Laboratory and were kept in an ærated aquarium. On a close examination of these colonies after a few hours, nearly all of the chitinous tubes were found empty, only a few containing the putrefying remains of the animal body. When I reexamined the colonies in the

aquarium on the next morning, I found that some younger animals had attached themselves to the tip or inner-side of the tubes of the departed generation; no doubt they had been hiding themselves somewhere in the colonial masses on the previous day. Most of these young worms measured  $1-2 \ c.m.$  in length.

I had the same experience in the summer of 1899. On August 2nd. I visited the place and dived under the ledge of rock where the colonies had formerly flourished; but I could obtain nothing but some decaying masses of the tubes which emitted a disgusting odour.

Judging from these facts, it seems to me not improbable that *Phoronis* annually changes its generation.

As to the formation of colonies of *Phoronis*, it may be supposed that the putrescent remains or a certain fluid secreted by the adult act on the larvae as a chemotropic reagent. But this can searcely be admitted as taking place in the wide and open sea. I think, on the contrary, that this phenomenon is not to be attributed to such a complex cause, but is to be regarded merely as an accidental matter. The colonies of Phoronis ijimai form a compact and rigid mass together with some Ascidians and Molluscan shells, and adhere very tightly to the rocks; so that, when once the animals form a colony in a suitable place, it may well be assumed that they become gradually luxuriant. But this is not really the case in Abaratsubo where the colony has remained almost the same in size for several years. I think, what takes place must be somewhat as follows: the places where the *Phoronis* colonies are established year after year, must naturally be well adapted to the life conditions of the worm, and when a large number of larvæ is metamorphosed, as must be the case, during the above mentioned months, those larvæ that happen to attach

582 1. IKEDA:

themselves to the tubes of the already formed colonies, flourish and attain full growth; on the other hand, if the larvæ become attached at some unfavorable places, they must soon be washed off by waves and many of them must perish before they can find other suitable places. To this wasteful death of the larvæ which have lost the opportunity of finding suitable localities to grow on, must be due the fact that they remain comparatively stationary in the number of colonies and in their distribution.

Specific Position of Phoronis ijimai Oka. According to Cori's table, there are 7 known species of *Phoronis*. But I can not refrain from entertaining serious doubts as to the correctness of the present mode of classifying the *Phoronida* in general. Most of the systematic data have hitherto been taken from the external characters of the animals, such, for instance, as the colour and size of the body, the number of tentacles, the general form of the colony, etc. The question now is whether these external characters are constant and can be depended upon for systematic use. there not a necessity for taking internal anatomical points into our consideration? According to my observations, Phoronis annually changes its generation and about one half of the year belongs to the growing period. Specimens collected during this growing season must necessarily differ from the adult in the breeding season in the number of tentacles, in the length and size of the body, etc. Wide discrepancies are therefore found between Oka's observation and mine on the same species, viz., Phoronis ijimai. I have no doubt that Oka made use of only the younger individuals as will be obvious from the following comparison:

	Body length.	Number of tentacles.	Length of tentacles.
Oka	$40 \ mm.$	150 (aver.)	2 mm.
Ikeda	$60-100 \ mm$ .	200-210 (aver.)	$\tilde{\mathfrak{I}}$ $mm$ .

1 think it possible that the distinction made between other species may rest on a similarly unsound basis.

Moreover there is another no-less important point to be considered. Soon after the report on *Phoronis buski* by Mc'Intosu ('88) was issued, Blandand Benham published his paper ('88) on the anatomy of *Phoronis australis*, in which he ascertained and rectified many important points which had been till then but incompletely known. Among these the following two are the most remarkable.

- (1) Afferent and efferent blood vessels open respectively into the recipient and the distributing vessels which run parallel and form so-called ring vessel. Each tentacular vessel is connected at its basal end not only with the recipient but also with the distributing vessel.
- (2) Each nephridial tube communicates internally with the infraseptal cavities by means of *two* funnels. One, the smaller, opens into the lateral chamber, while the second is considerably larger and opens into the rectal chamber.

It is stated by Benham, that "Mr. Caldwell dealt only with the larger of the two funnels, in his 'preliminary note.' but he informed me by letter that he became aware of the existence of the second funnel, shortly after the publication of his paper.' How is it now with the ring vessel in *P. kowalewsky*? Caldwell says nothing about it. Corl, on the contrary, denied the above characters for *P. psammophila*.

In *Phoronis ijimai* I have ascertained that these two structures are the same in every point as in P, australis. Fig. 62 a shows a dorsal portion of a transverse section through the septum; two nephridial canals (nep.e.) are seen one on each side of the intestine (int.) and are partly imbedded in the chondroid tissue of

the septum (mes.). These canals open by funnels (f.) into the body-cavity (rectal chamber) which separates the intestine (int.) from the esophagus (æs.). These funnels correspond to the larger funnels of Benham, so that in a longitudinal section they appear as long ciliated cell-masses running longitudinally along the lateral mesenteries. If we trace the funnels a little downwards, we find another kind of funnels on the opposite sides of the lateral mesenteries, opening into the lateral chambers. They are represented in fig. 62 b (f.'), which is taken from the right nephridium. They are short indeed and are apt to be overlooked.

Again as to the ring vessels, they are found always as two concentric loops (the recipient and the distributing) standing side by side, and, as was described by Benham, each tentacular vessel receives two small branches respectively from the two vessels. Besides, in *Phoronis hippocrepia*<sup>th</sup> which is known from Ilfracombe, I have ascertained that the above cited structures (the nephridia and the ring vessels) are indubitably present without any modification.

If these structures can not really be found in *Phoronis psam-mophila*, similar specific anatomical deviations must exist in other little studied species, for instance, in *P. ovalis*, *P. gracilis*, *P. buski*, and so on. From these facts and from the variability of the external characters, I am at present unable to discover points by which *Phoronis australis*, *P. hippocrepia*, and our *Phoronis* can be differentially diagnosed.

Tokyo Imperial University, Science College.

October, 1899.

<sup>&</sup>quot;For the opportunity of investigating this species, I am much indebted to Prof. Yasuda of the Second Higher School who kindly gave me a small portion of a colony.

## Postscript.

While the manuscript of the present article was undergoing revision, I was much pleased to read Roule's elaborate work on the development of the *Phoronida*.\* For the sake of brevity I will not here discuss the author's theoretical considerations, but will offer some remarks with regard only to his investigations relating to developmental facts. Some of his results differ considerably from mine and from those of previous observers. And the differences are such as do not seem to be due merely to different conditions in the species investigated (*Phoronis sabaticri*).

According to Roule's observations, the first four planes of segmentation are all vertical and radial, the fifth being the first that is horinzontal, thus giving rise to 16 blastomeres; and the egg is composed of two sorts of larger and smaller cells from the first cleavage. This differs considerably from the account given in the preceding pages, which is in agreement with the studies of Foettinger and Masterman (1900). It is, however, very difficult to decide which of the two opinions is correct or whether both are correct. I am at present rather inclined to the latter view. As to Roule's belief in the peculiar unequal segmentation, which is said to return soon after to the equal, I fear that the eggs dealt with by the author may have been somewhat premature. I have often observed premature eggs undergoing a remarkable unequal segmentation when mixed in water with spermatozon.

With respect to the nature of plasmic corpuscles, Roule's view is certainly identical with that of Caldwell, though he does

<sup>\*</sup> Etude sur le development embryonaire des Phoronidiens.—Ann. d. Sci. Nat. Zool., T.XI . No. 1-6, 1900.

not refer to the latter author. In examining Roule's figures (31 and 32), certain nucleated cells are seen dispersed in the blastocelic cavity. They are said to be the inner ends of some elongated blastomeres. In the embryos of *Phoronis ijimai*, similar processes are indeed discovered protruded from some blastomeres, but they nerver contain the nucleus in their distal or inner ends: the nucleus belonging to these elongated blastomeres is situated also peripherally as in other normal blastomeres. The plasmic corpuseles which have, as I have described in the present paper, arisen from subsequent fragmentation of certain elongated blastomeres, exist very distinctly as separate bodies dispersed in the blastocelic cavity.

In his present paper ROULE reiterates his former views about the dual origin of the mesenchyme-cells. Upon this point I have already given my own ideas, and here I have to add only the following remark:

Though ROULE, like SCHULTZE ('97) has regarded, the nephridial pit as the origin of the ventral pouch of Actinotrocha, the subsequent development distinctly shows that the two structures are entirely independent of each other in their origin.

I can not refrain from doubting the correctness of Roule's observation that the larva studied by him possessed at no time during the swimming life any septal membrane in the body-cavities. They are structures which in the other forms of Actinotrocha have been so accurately demonstrated by previous observers. The technique employed by the author may perhaps be found to be faulty in this respect. The fine threads denoted by "brides mesenteriques" in his figures (57, 75, and 97) seem to have arisen from the pieces of the otherwise continuous postral septum broken by the knife-blade in microtomizing.

Roule has described the nephridia of the larva in a somewhat peculiar way. According to him, the organs lie considerably anteriorly as they are found on both sides of the stomach diverticulum, and are said to be constructed of cells forming a syncytial mass which is attached to the somatic walls. No lumen and no leading canal have been detected in these masses. But, if judged from the author's text and figures, it seems to me highly probable that his so-called nephridia correspond to the corpuscle masses of other writers. Having overlooked the postoral septum and the true corpuscle masses, he seems to have come to mistake the latter for nephridia. Thus, so far as I can understand his description, he did not notice the change in position of the organs with regard to the postoral septum during metamorphosis.

As to the number and the position of the stomach diverticulum, the larva of *Phoronis sabatieri* is described to be in the same condition as that of *P. ijimai*. But the vacuolization process of the organ is in the former species more complex than in the latter, though it is simpler than in the larvae studied by Masterman. When these three cases are considered together, it may be concluded that they indicate specific variations.

Roule's "cordon dorsal," which is considered to show the rudimentary state of the rectum of the adult, has not been described by any previous author, and also could not be detected in any of the types of the larvæ studied by me. If this "cordon dorsal" be reconstructed from Roule's text and figures, it seems to me almost without doubt that the structure referred to corresponds to the dorsal vessel on the stomach. When Roule's figure (75) and mine (58c) are compared, both of which show a cross section through the tentacular region at a similar level, it will be noticed that the "cordon dorsal" and the "vaisseau dorsal" in

the former figure correspond respectively to the dorsal vessel and the trunk cavity (anterior portion) in the latter. Again, in regard to the alimentary canal, Roule states that the end portion of the intestine atrophies, but according to the observations of Caldwell and myself, no portion of the larval alimentary canal, except the stomach diverticulum, undergoes histolysis during metamorphosis, the entire tract growing gradually in length.

Roule's views as to the origin of the blood vessels greatly differ from those of Caldwell, Masterman and myself. According to Roule, the vascular spaces and the body-cavities are ontogenetically the same thing, and the former is formed from a coalescence of the irregular lacunal spaces of the latter. Though I agree with him in considering the ring vessel of the adult as a derivative of the body-cavity (collar) of Actinotrocha, yet I can not accept his view attributing other vessels to the same process. Besides, we see in his figures (82, 83, 86, 87, 88, etc.) that the same vessel (dorsal) is placed sometimes between the afferent, and the efferent, branches of the intestine, and sometimes on the somatic walls. Can such a peculiar disposition of the blood vessels against the skin be verified in the adult animal?

It may be known from Roule's contributions, that the larval development is more accelerated in *P. sabatieri* than in other species. So that the ventral pouch and some other organs are in that species already well developed even in a larva of 6 tentacles. But this seems of not much significance, since we know that even in the same type of the larvae the progress of larval organisation does not always keep pace with the increase in the number of the tentacles.

Finally I can but refer here to MASTERMAN's new work (1900),\* which I was able to read only a short time ago. His views differ so radically from those of previous authors and from my own, that I cannot fully discuss so weighty a matter in a brief postscript.

May, 1901.



<sup>\*</sup>On the Diplochorda, III, the early development and anatomy of Phoronis Buskii, McI. Quat. Jour. Micr. Sci., Vol. 43, 1900.

## Reference papers.

- '46. J. MÜLLER.—Bericht über einige neue Thierformen der Nordsee (Müller's Arch. f. Anat. u. Phys., 1846).
- '47. R. Wagener.—Actinotrocha branchiata (ibid., 1847).
- \*54. C. Gegenbaur.—Bermerkungen über Pilidium, Actinotrocha, etc. (Zeitsch. f. Wiss. Zool., Bd. V. 1854).
- \*54. A. Krohn.—Ueber Pilidium and Actinotrocha (Müller's Arch. f. Anat. u. phys., 1854).
- '62. A. Schneider Ueber die Metamorphose von Actinotrocha branchiata (ibid., 1862).
- '71. E. Metschnikoff.—Ueber die Metamorphose einiger Seethiere (Zeitsehr, f. Wiss. Zool., Bd. 21, 1871).
- '80. E. B. Wilson.—On Actinotrocha (Amer. Natur., 1880).
- '81. E. B. Wilson.—The origin and significance of the metamorphosis of Actinotrocha (Quart. Journ. of Microsc. Sci., Vol. XXI, 1887).
- '82. E. Metschnikoff.—Vergleichende embryologische Studien über die Gastrula einiger Seethiere (Zeitschr. f. Wiss, Zool., Bd. 27, 1882).
- '82. A. Foettinger.—Note sur la formation du mésoderme dans la larve du Phoronis hippocrepia (Arch. d. Biolog., Tom. 111, 1882).
- \*82. H. Caldwell.—Preliminary note on the structure, development, and affinities of Phoronis (Proc. R. S. L., Vol. 34, 1882-1883).
- '85. H. Caldwell.—Blastopore, Mesoderm, and Metameric Segmentation (Quart. Jour. Micro. Sci., Vol. 25, 1885).
- '88. Mc'Intosii.—Report on Phoronis Buskii (Challenger Report, XX, part LXIII, 1888).
- \*88. B. Benham.—The anatomy of Phoronis australis (Quart. Jour. Micr. Sci., Vol. XXX, 1888).
- '90. L. Roule.—Sur le développement des feuillets blastodermiques chez les Gephyriens tubicoles (C. R. Ac. Sci., CX, 4890).
- '96. L. Roule.—Sur les metamorphoses larvaire du Phoronis sabatieri (ibid., 1896).
- '97. Λ. Οκλ.—Sur une nouvelle espèce japonaise du genre Phoronis (Annot, Zool, Japo., Vol. 1, 1897).

- '97. E. SCHULTZE.—Ueber Mesodermbildung bei Phoronis (Trav. Soc. Imp. Sci. Nat. St. Petersburg, Vol. XXVIII, 4897).
- '97. A. T. Masterman.—Preliminary note on the structure of Phoronis (Proc. Roy. Soc. Edinb., Vol. XXI, 1897).
- '97. A. T. Masterman.—On the structure of Actinotrocha considered in relation to the suggested Chordate affinities of Phoronis (ibid., Vol. XXI, 1897).
- \*97. A. T. Masterman.—On the Diplochorda (Quart. Jour. Micr. Sci., Vol. XL. 1897).
- '98. L. Roule.—Sur la place des Phoronidiens dans la classification des animaux et sur leur relations avec les Vertebrés (C. R. Ac. Sci., 1898).

#### Postscript.

'99. L. Roule.—La structure de la larve Actinotroque des Phoronidiens (Proc. 4th Intern. Congr. Zool., 1899).

### List of Abbreviations.

un., anus. ant.div., auterior diverticulum. a.v., afferent vessel. bl., blastopore. bl.c., blastoccelic pore. cir.c., ring vessel, i.e., the collar cavity. col, collar of larva. col.c., collar cavity. corp.. blood corpuscles and corpuscle mass. dig.a., digestive area. div., stomach diverticulum. d.v., dorsal vessel. ect., ectoblast. e.rc.c., excretory cells. f. and f'. nephridial funnels. t.n., female pronucleus. gl., nerve ganglion. gld., gland. int.. intestine. m., mouth. mes., postoral septum. mes'., preoral septum. Mes., mesoblast cells. m.ql., mucous gland. m.f., mesenchymatous fibres. m.n., male pronucleus. m.sh., muscular sheath of the stomach. nep.c., nephridial canal. neph., nephridium. nep.o., nephridial pore.

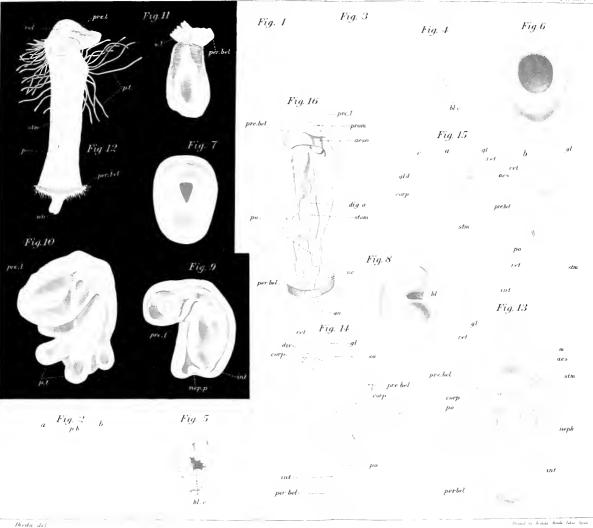
nep.p., nephridial pit. n.f., nerve fibre. as., esophagus. o.po., pouch pore. p.b., polar globule. p.c.c., larval collar cavity. per., peritoneal epithelium. per.bel., perianal belt. pl.co., plasmic corpuscle. po., ventral pouch. pre.bel., preoral belt. pre.c., preoral body-cavity. pre./., preoral lobe. p.r., posterior recess of preoral cavity. p.t., larval tentacle. ret., retractor muscle in the collar. ret'., retractor muscle in the trunk. s.c.c., adult collar cavity. s.o., sense organ. s.s., sinus space. s.t., adult tentacle. stm., stomach. t'., t''., t'''.........first, second, third larval tentacles. tr., trunk. tr.e., trunk cavity. t.v., tentacular vessel. v.c., vascular ceca or capillaries.  $v_{eff}$ ., ventral groove. v.mes. ventral mescutery. v.v., ventral vessel.

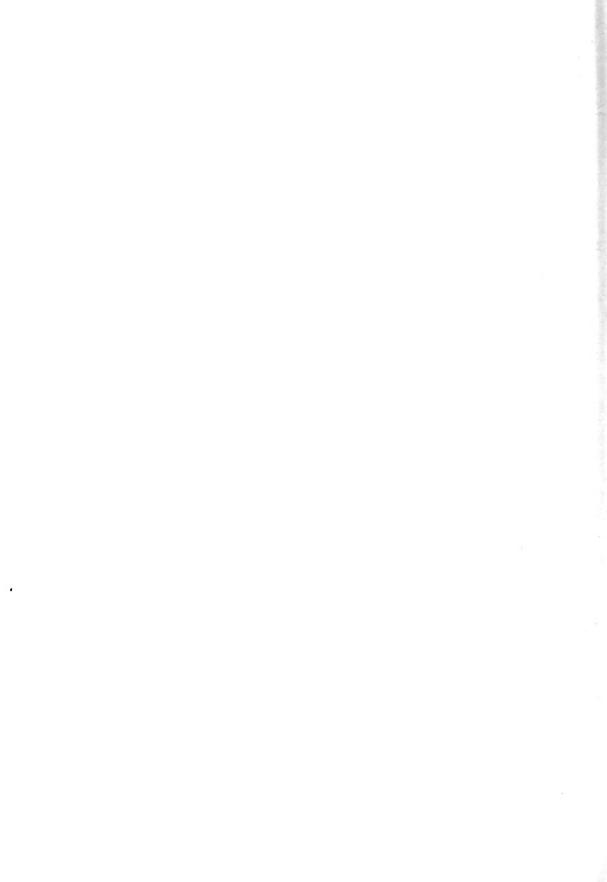


# Explanation of Figures.

## Plate XXV.

- Fig. 1.—Egg with two blastomeres. ×4 B (Zeiss).
- Fig. 2.—Egg with three (a) and four blastomeres (b).  $\times$  4 B.
- Fig. 3.—Egg with eight blastomeres, side view.  $\times$  4 B.
- Fig. 4.—Egg with thirty two blastomeres, seen from the future ventral side.  $\times$  4 B.
- Fig. 5.—Young morula, ventral view.  $\times$  2 D.
- Fig. 6.—Ventral view of an advanced blastula in which the gastral invagination has become visible from the outside.  $\times$  2 D.
- Fig. 7.—Ventral view of a gastrula, in which the blastopore has taken a triangular form.  $\times$  2 D.
- Fig. 8.—Side view of an advanced gastrula, in which the blastopore has become a transverse slit.  $\times$  2 D.
- Fig. 9.—Young Actinotrocha in which the nephridial pit is visible from the outside.  $\times$  2 D.
- Fig. 10.—Larva of four tentacles (side view).  $\times$  2 D.
- Fig. 11.—Metamorphosing larva of type A, sketched from a preserved specimen.  $\times$  4 A.
- Fig. 12.—Larva of type D, sketched from a living specimen. Greatly magnified.
- Fig. 13.—Larva (of 14 tentacles) of type A, in the living state. Greatly magnified.
- Fig. 14.—Highly advanced larva of type B, bearing 28 tentacles, ventral view. Greatly magnified.
- Fig. 15.—Larva of type C, bearing 20 tentacles. a represents a dorsal view, b a ventral view, and c the multicellular gland on the hood. Greatly magnified.
- Fig. 16.—Larva of type D, bearing 48 tentacles, after preservation. Greatly magnified.

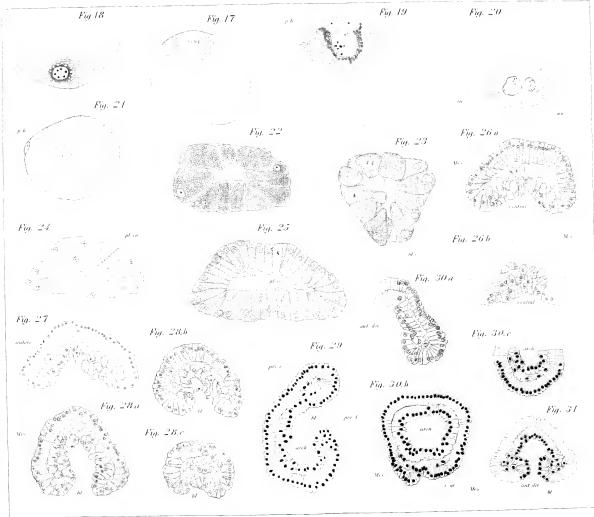






## Plate XXVI.

- Fig. 17.—Primary occyte, showing the karyokinetic figure for the first polar globule.  $\times 2$  imm.  $^{-1}/_{12}$ .
- Fig. 18.—Section through the equatorial plane of the karyokinetic figure for the first polar globule; the egg was preserved with sublimate solution (Winkel oc, 3.  $\times$  ob, 8).
- Fig. 19.—After emission of two polar globules (Winkel oc.  $3 \times ob$ , 8).
- Fig. 20.—Section showing one stage of fertilization, when the two pronuclei (m.n. and f.n.) stand side by side.  $\times 2 \text{ imm.}^{-1}/_{12}$  (Zeiss).
- Fig. 21.—Median section of fertilized egg, in which is found a karyokinetic figure for the first cleavage. × 2 imm.  $\mathbb{V}_{p}$ .
- Fig. 22.—Median section of a young mornla.  $\times$  2 F.
- Fig. 23.—Median section of a young morula, showing the blastocolic pore (bl.c.).  $\times$  2 F.
- Fig. 24.—Median section of a young blastula, in which one blastoderm cell is seen giving off plasmic corpuscle (pl.co.).  $\times$  2 F.
- Fig. 25.—Median sagittal section of an advanced blastula; two plasmic corpuscles are detected in the segmentation cavity. × 2 F.
- Fig. 26 (a, b).—Sagittal sections of a highly developed blastula, in which the invagination has just begun; a shows a median section.  $\times$  2 imm.  $^{1}/_{10}$ .
- Fig. 27.—Median sagittal section of a gastrula in which the invagination is deeper.  $\times$  2 imm.  $^{1}/_{12}$ .
- Figs. 28  $(a \cdot c)$ .—Three transverse sections of an advanced gastrula : a through the central depression, b behind the central depression, and c near the posterior end of the embryo.  $\times$  2 F.
- Fig. 29.—Median sagittal section of an embryo at nearly the same stage as in Fig. 8.  $\times$  2 imm.  $\frac{1}{12}$ .
- Figs. 30 (a cdot c).—Transverse sections of an embryo at nearly the same stage as the preceding; a shows a portion (left) of a section through the blastopore, b just behind the blastopore and through the ventral groove (v.gr.), c near the posterior end.  $\times$  2 F. a with the tube drawn out.
- Fig. 31.—Transverse section through the blastopore of a larva, in which the anterior diverticula (ant, div.) are well developed.  $\times$  2 F.

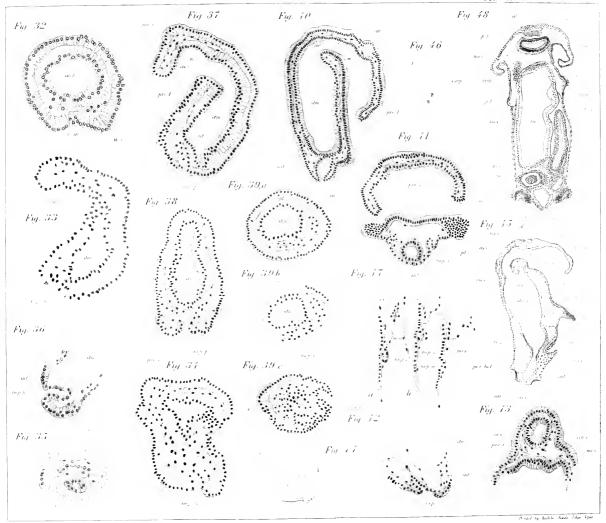


			,	



### Plate XXVII.

- Fig. 32.—Transverse section through the middle portion of a larva in which the ventral groove has ceased to give off mesoblast cells.  $\times$  2 F, with the tube drawn out.
- Fig. 33. Slightly oblique sagittal section of a larva in which the nephridial pit (uep.p.) has made its first appearance.  $\times$  2 imm.  $^{1}/_{12}$ .
- Fig. 34.—Oblique frontal section of a larva nearly at the same stage as in Fig. 9.  $\times$  2 F.
- Fig. 35.—Transverse section through the nephridial pouch (nep.p.) as yet unpaired.  $\times$  2 F.
- Fig. 36.—Oblique sagittal section of the nephridial pouch (nep.p.) partly divided.  $\times$  2 F.
- Fig. 37 and Fig. 38.—Show respectively sagittal and frontal sections of larvæ, in which the proximal end of the nephridial pit is about to divide into two. × 2 F.
- Figs. 39 (a-c).—Three transverse sections of the two nephridial canals, each of which has respectively an internal opening.  $\times$  2 F.
- Fig. 40.—Sagittal section of a larva at the stage represented in Fig. 10.  $\times$  2 F.
- Fig. 41.—Frontal section of a larva at the same stage as the preceding, in which the two nephridial pores have separated widely from each other.  $\times$  2 F.
- Fig. 42.—Oblique frontal section through the nephridial region of a larva a little younger than the preceding.  $\times$  2 F.
- Fig. 43.—Transverse section through the upper portion of the cosophagus of a larva of four tentacles.  $\times$  2 F.
- Fig. 44.—Large mesoblast cells which are found in the body-cavity of the larva of two or four tentacles.  $\times 2$  F.
  - (All the figures from fig. 45 to fig. 55 are drawn from larve of type A.)
- Fig. 45.—Median sagittal section of a larva of 12 tentacles.  $\times$  2 D.
- Fig. 46.—Portion of a longitudinal section of a larval tentacle of 10-armed larva; two blood corpuscles are represented in the tentacular cavity (corp.).  $\times$  2 F.
- Figs. 47 (a-c).—Three longitudinal sections of the nephridium of a larva of 12 tentacles,  $\times$  2 F.
- Fig. 45,-Median frontal section of a knya of 16 tentacles,  $\times$  2 D.



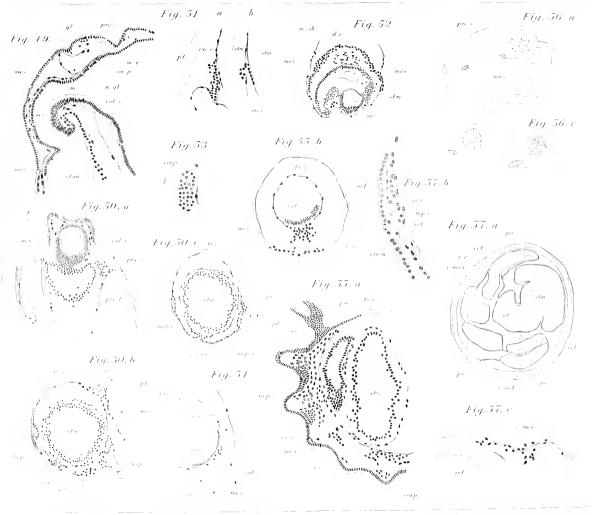


## Plate XXVIII.

- Fig. 49.—Anterior portion of a median sagittal section of a larva of 14 tentacles,  $\times$  2 F.
- Figs. 50 (a-c).—Three transverse sections of a larva of 14 tentacles; a through the stomach diverticulum (div.), b obliquely through the postoral septum (mes.), and c above the pouch pore.  $\times$  2 D.
- Figs. 51 (a, b).—Longitudinal sections of the nephridium of a larva of 16 tentacles.  $\times$  2 F.
- Fig. 52.—Transverse section through the junction of the stomach and the cesophagus of a larva of 16 tentacles, showing the radiment of the dorsal vessel  $(d.c.) \times 2$  imm.  $\frac{1}{1/12}$ .
- Figs. 53.—Section of the above larva, through the corpuscular mass which thoats in the collar cavity.  $\times$  2 F.
- Fig. 54.—Sagittal section through the right side of the asophagus of a larva of 16 tentacles.  $\times$  2 F.
- Figs. 55 (a and b).—Cross sections of a larva of 16 tentacles. In the figure a is represented the right half of the section which passes through the stomach (stm.) and the adult collar cavity (s.c.c.): b through the junction of the stomach and the intestine, whereto the contractile ecca (v.c.) are attached.  $\times$  2 F.

# (Figs. 56-57 are drawn from larva belonging to type ('.)

- Figs. 56 (a-c).—Cross sections of the hood, where the multicellular gland is represented in different planes,  $\times$  2 D. Letter b omitted.
- Figs. 57 (a-c).—Cross sections through the trunk of a larva of 22 tentacles: a under the magnification of  $2 \times D$ ; b a portion of the trunk walls containing the nephridial canal, magnified  $2 \times F$  with the tube out; c shows a portion of the ventral mesentery near the gut, magnified  $2 \times F$ .







# Plate XXIX.

(Figs. 58-60 are drawn from larve belonging to type D).

- Fig. 58 (a-e).—These figures show different parts of a larva of 44 tentacles. Figure a shows a cross section through the mouth, b through the middle portion of the æsophagus (as.); magnified  $\times$  2 B; c a portion of a cross section of the stomach wall (dorsal) and the trunk walls,  $\times$  2 F. d and e show respectively a longitudinal and a transverse section of a tentacle (the former magnified  $\times$  2 D, the latter  $\times$  4 D).
- Figs. 59 (a-d).—Four cross sections taken from a series, not consecutive, and their respective planes of section are given in the text with reference to the woodcut (p. 542). Unfortunately in these series, the tissues have undergone a great disturbance by the killing reagent, but the relations of the layers remain essentially correct, and those spaces which have been produced from the mutual splitting of the layers are denoted by "artefact." × 2 D.
- Figs. 60 (a and b).—Represent the nervous system of Actinotrocha (of type B), revealed by vital staining with methyl blue and ammonium molybdate. In a, as the larva was pressed by the coverglass, the rim which appears like the free margin of the hood is not that edge at all, but represents the line along which the hood was bent by pressure; the line drawn near the peripheral blue dots is the true edge of the hood. b shows a portion of the free margin of the hood, where the nerve fibres end.  $a \times 4$  B,  $b \times 2$  F.
- Figs. 61 (a and b).—Are taken from serial sections of a A-type larva which bears the evaginated pouch; a shows four blood corpuseles, b one portion of the wall of the stomach diverticulum.  $\times$  2 F.
- Figs. 62 (a and b).—Taken from serial transverse sections through the nephridial region of the adult. b shows the inner and of the left nephridial canal where the smaller funnel (j") is attached.

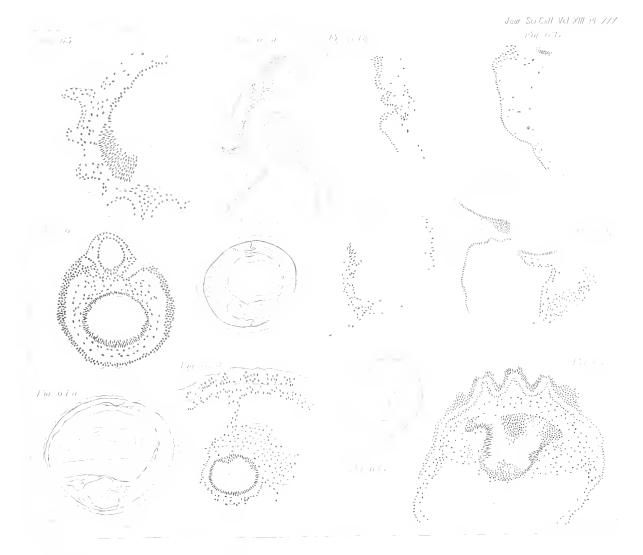






#### Plate XXX.

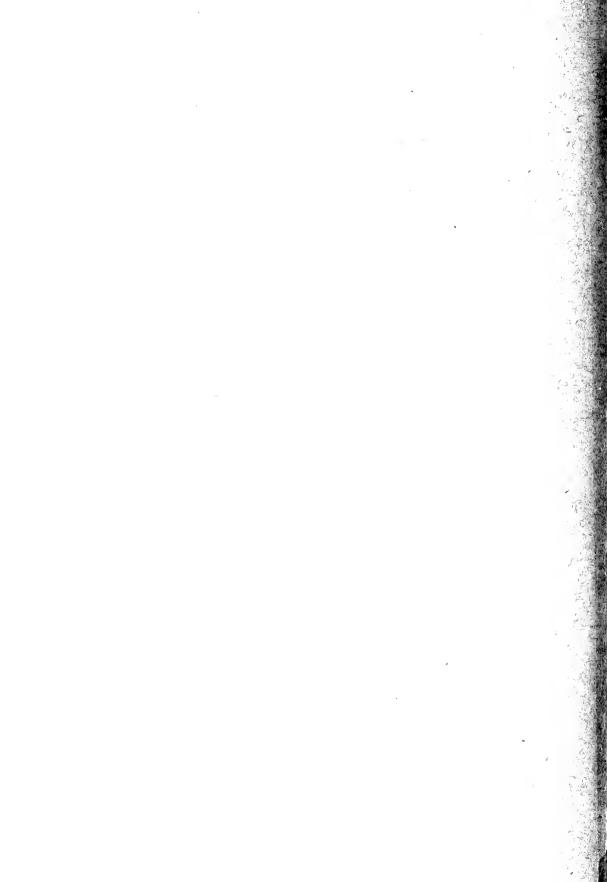
- Figs. 63 (a-e).—Are taken from several parts of serial sagittal sections of B-type larva of 28 tentacles.
  - a. median section through the hood and the collar.  $\times$  2 D.
  - b. more magnified figure of the nerve ganglion (gl.) and the posterior recess of the preoral cavity (p.r.) in the preceding figure.  $\times$  2 F.
  - c, taken from a section lateral to the assophagus and to the right of that of the figure a.  $\times$  2 F.
  - d. ventral portion of the collar-trunk walls, where the septum (mes.) and the adult collar cavity (s.c.c.) are cut through.  $\times$  2 F.
  - e. portion of a section which passes through the second tentacle (t'').  $\times$  2 F.
- Figs. 64 (a-f).—Transverse sections taken from serial sections of a metamorphosing larva of type A represented in fig. 11. The respective explanations of them are introduced in the text (p, 584). From a to c magnified as  $\times 2$  D, and from d to f as  $\times 2$  F.
- Fig. 65.—Portion (right side) of a transverse section through the tentacular region of a metamorphosing larva of type A, slightly younger than that of fig. 11.  $\times$  2 F.
- Fig. 66.—Transverse section through the nephridial region of wholly metamorphosed larva of type A.  $\times$  2 F.



J.

01





WH 19K6 %